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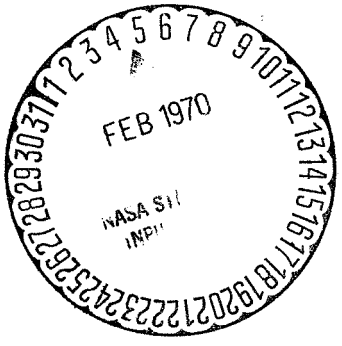
VOYAGER MARS PLANETARY QUARANTINE: COLD GAS ATTITUDE CONTROL SYSTEMS, EXPERIMENT PROGRAM - FINAL REPORT, MARCH 1967



JET PROPULSION LABORATORY
CALIFORNIA INSTITUTE OF TECHNOLOGY
PASADENA, CALIFORNIA

DOCUMENT NO. VOY-C2-TR9
MARCH, 1967

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PREPARED BY:

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PROPULSION ENGINEERING
ADVANCED INTERPLANETARY
PROGRAMS

APPROVED:

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R. P. WOLFSON, COGNIZANT
ENGINEER
PLANETARY QUARANTINE
VOYAGER SPACECRAFT SYSTEM
PROJECT

PREPARED FOR

JET PROPULSION LABORATORY
CALIFORNIA INSTITUTE OF TECHNOLOGY
4800 OAK GROVE DRIVE
PASADENA, CALIFORNIA

UNDER JPL CONTRACT NO. 951112

GENERAL ELECTRIC
MISSILE AND SPACE DIVISION
Valley Forge Space Technology Center
P.O. Box 8555 • Philadelphia 1, Penna.

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ACKNOWLEDGEMENT

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C. C. Rich, R. T. McFall
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M. G. Koesterer. R. C. Bateson
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SECTION 1

SUMMARY

This report presents the work performed in determining the biological loading of a cold gas attitude control system ejecta in support of the Voyager Task C Planetary Quarantine Study Program. The task was divided into two phases, a literature search, and a test program. The literature search was completed in October 1966 and was reported in Appendix F. The literature search provided the following information:

a. Attitude control pneumatic systems of the size considered for Voyager could contain from 10^4 to 10^5 viable microorganisms on the interior surfaces after being cleaned according to the listed cleaning specifications. (See Appendix F.)

b. Purging components with sterile nitrogen was a relatively ineffective method for removal of biological contamination as shown by the test conducted at GE.

c. Filtration gave indications of being an effective and practical means of removing biological contamination from gases.

d. Ethylene oxide (ETO) decontamination and dry heat sterilization are capable of reducing the biological loading by 10^8 and 10^{12} resistant organisms, respectively. However, the heat cycle and ETO process may have detrimental effects on component life and reliability.

From information gained in the literature search, a test program was justified and immediately initiated to determine the number of viable organisms that would be discharged from an attitude control system that was cleaned and assembled according to current state-of-the-art processes. A breadboard single-axis cold gas system was designed, utilizing typical spacecraft (ACS) hardware and possessing proportional internal area (pressurant tank to rest of system) and being functionally similar to that of the GE Task B proposed ACS. This breadboard system was cleaned according to the Nimbus spacecraft specifications and assembled in a Class 100,000 clean room. The system was pressurized with biofiltered nitrogen gas to 2000 psi and activated on a duty cycle consisting of a series of 300-millisecond, 5-second, and partial blowdown firings.

During the testing, the tank pressure, regulated pressure, and the nozzle chamber pressure were recorded on an oscillograph on a time base. The discharge gas from each firing was filtered by means of exhausting it through a 0.45-micron membrane filter in a high-pressure filter holder. The filters were bioassayed after the test to determine the number of organisms recovered. Results of the bioassay are presented in Tables 8-2 through 8-9.

Five tests were performed with the breadboard system, of which two had a vibration environment prior to testing, two were tested as assembled, and one was a controlled contamination test. A NIMCO system (Nimbus Satellite, engineering model) was also tested during this effort. Bioassay results of the filters used in the vibration and as-assembled breadboard system test show viable spores existing on nine of 57 filters cultured for a total valid spore colony count of 52. A total of 34 ft^3 of gas @ 1 atm was expelled in those four tests. The average spore colony discharge rate was 1.52 spores/ft^3 of gas. The NIMCO system test provided 2.85 spore colonies/ft^3 of gas. These contamination values can be considered as maximum, because the background contamination level (contamination introduced during the bioassay procedure) at times was equal to that obtained from the ACS firings.

To ascertain the system internal surface contamination level, the in-line filter of the breadboard system was bioassayed. No contamination was recovered. Four additional components, two filters and two solenoid values, were also bioassayed. These components were not used in the breadboard system but are of the same part number as comparable parts in the system. A filter and a solenoid value were purged clean to the Nimbus particulate cleanliness level, and a filter and solenoid valve were ultrasonically degreased in a Freon bath and then purged cleaned to the Nimbus requirements. The bioassay results show that the ultrasonically cleaned filter and solenoid valve had 75 and 0 spores recovered, respectively, while the purge-cleaned components had a biological contamination level that was so high a count could not be taken.

SECTION 2

INTRODUCTION

The study to determine the effects of propulsion systems on the maintenance of Mars planetary quarantine is being conducted as part of the Voyager Task C Planetary Quaratine Program. A potential source of contamination is the release of viable organisms from the spacecraft cold gas attitude control system.

The biological discharge values obtained from this study are to be utilized with other ACS parameters (gas discharge velocity, direction, rate, etc.), within a math model, formulated to assess the probability of a single viable organism reaching Mars.

SECTION 3

PROGRAM PLAN

The cold gas biological contamination investigation was conducted as follows:

a. A literature search to investigate previously conducted cold gas system biological control efforts.

b. Performance of biological assay, testing, and analysis where previous investigations were deficient in satisfying the quarantine study needs.

SECTION 4
LITERATURE SEARCH

The literature search was completed in October'66 and reported in Appendix F. The following items are given as a summary of the report.

a. As part of a NASA contract,* GE performed a test to determine the bioloading of hardware components. In this test, the interior surfaces of three sets of pneumatic components were bioassayed after the components were cleaned to specific cleanliness procedures. The results show that the components would contain approximately 200 viable organisms/ft^2. However, in the testing it was determined that the bioassay technique was, as a minimum,10 percent efficient, and therefore placed the actual component bioloading at a maximum of approximately 2000 organisms/ft^2.

b. Under the same contract,* it was found that purging by sterile nitrogen gas or water was a relatively ineffective means of removing organisms from internal surfaces of pneumatic hardware components.

c. Heat sterilization and decontamination by ETO when applied in an effective manner would reduce the viable organism loading on surfaces by 10^{12} and 10^{8} organisms, respectively. However, little is known about the effect on component reliability by these decontamination processes. Heat sterilization appears to provide the greatest unknown and probably the most severe restriction on system and component design. ETO does not appear to present as large a compatibility problem as heat sterilization. ETO is not considered a sterilizing process where bioassay of a surface cannot be performed, i.e., internal surfaces.

d. Gases and liquid can be sterilized by filtration. Filter manufacturers have performed tests, under carefully controlled conditions, that have resulted in removing all particles larger than the absolute pore size of the membrane filter from liquids and gases.

*NASA Contract No. NAS 8-11372

SECTION 5

TEST PROGRAM

The test program was designed to provide results that could be used to predict, within reasonable accuracy, the quantity of viable organisms (spores) that would be ejected from a Voyager-size cold gas ACS. The following considerations were applied to the breadboard ACS design.

a. The ratio of propellant tank internal area to all other component and line internal areas is the same for the test system as for the Voyager ACS proposed by GE in the Task B design. This ratio is approximately 80/20 and represents the high-velocity and low-velocity gas areas.

b. The test system utilizes typical ACS pneumatic components to give representative internal areas, flow passage configurations, and gas velocities.

c. A thruster that provides the same system gas flow rate as any single thruster on the Voyager System.

d. A test cycle that consists of short-duration and long-duration pulses at intervals along the ACS operational spectrum.

e. Vibration was applied to the test system at the expected Voyager frequencies and accelerations prior to system operation. This test is to determine the effect of vibration on bio-ejection rate.

f. The system was pressurized with biofiltered gas (filtered through 0.45-micron filters) so that all organisms subsequently ejected may be considered to have been removed from the internal surfaces of the system.

As a check on the validity of the data obtained from the breadboard system test, the ejecta of the NIMCO system was bioassayed. The NIMCO system,at the time of testing, was being prepared for another test program and was cleaned and assembled to the same specifications as the breadboard system. Access to the system for bioassay was limited because of the NIMCO test program, and only 1.4 ft^3 of gas (1 atm) was sampled. The gas was expelled from the positive roll and negative pitch nozzles on 5-second pulse durations. Twelve filters were used in the test, one for each nozzle per 5-second pulse.

The above work was performed by the GE Spacecraft Department in accordance with the work statement presented in Appendix B.

A controlled contamination test was also conducted where a known quantity of spores (Bacillus subtilis var niger) were injected into the system. This test was performed to determine the amount of biocontamination that would be ejected under known contamination conditions, and to place a value on the effectiveness of the bioassay techniques. This test was performed last to preserve the breadboard system cleanliness for the "as-built" evaluation testing.

To provide a correlation between internal surface contamination level and ejecta bioloading, the breadboard system "in-line" filter (assumed to be the component that would have the highest contamination level in the system) internal surfaces were bioassayed. Other system components were not bioassayed because the bioassay procedure, to be effective, would require a component which could result in destroying the components for further use in their intended capacity. However, to provide additional component internal contamination data, four components that were not used in the system were bioassayed after being cleaned according to the Nimbus specification.

The test program also included testing as required to determine system functional characteristics and to develop bioassay techniques.

SECTION 6

SYSTEM CONFIGURATION

A detailed description and layout drawing of the single axis breadboard system was given in the program status report, Appendix G. A system schematic and specific system parameters are given in Figure 6-1 and Table 6-1, respectively.

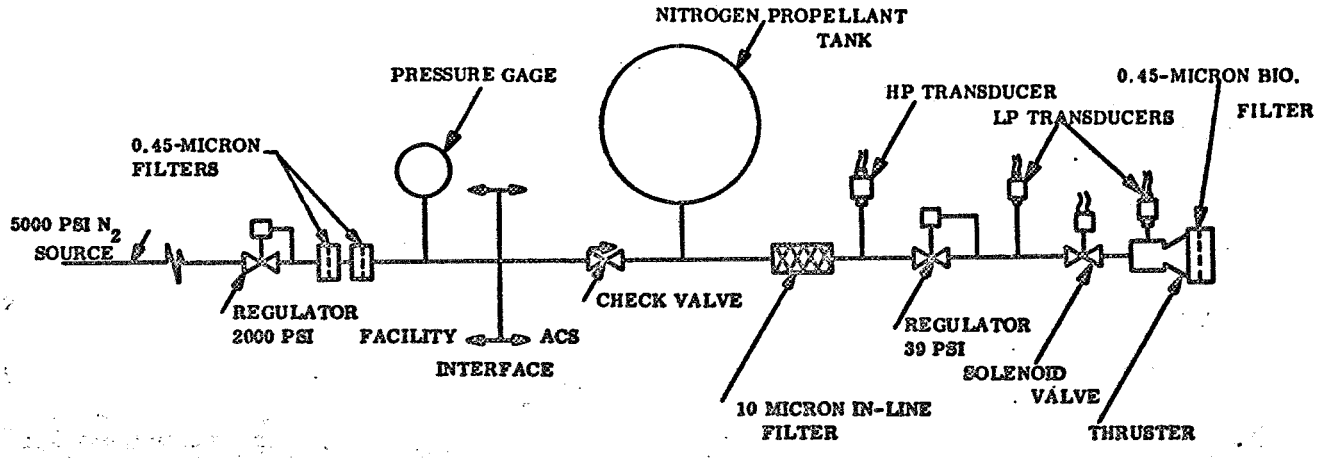


Figure 6-1. Breadboard Attitude Control System Schematic

Table 6-1. System Operating Parameters

Parameter	Value
Tank volume	142 in^3
Tank pressure	2000 psi
Weight of gas	0.815 pounds
Volume of gas	10.45 ft^3 at 1 atmosphere
Regulated pressure	39 psia
Thruster chamber pressure	36 psia, 51 psia
Gas flow rate	0.0019 lb/sec 0.0243 ft^3/sec
System thrust	0.1 pounds
System total internal Area	178.0 in^2
Tank to system area (percent)	79.5 percent

A schematic of the NIMCO system tested is given in Figure 6-2. The internal area of the NIMCO system was not readily computed. The difference in internal area between the breadboard and NIMCO systems exists primarily in the tankage and the tubing, since the remaining components are identical in many cases. The internal area of the NIMCO system as a single-axis system is approximately 400 percent larger than the breadboard system. The tank-internal-area-to-remaining-system-area ratio is 62 percent. The NIMCO operating parameters are the same as the breadboard system (i.e., thrust chamber pressure, regulated pressure, and gas flow rate).

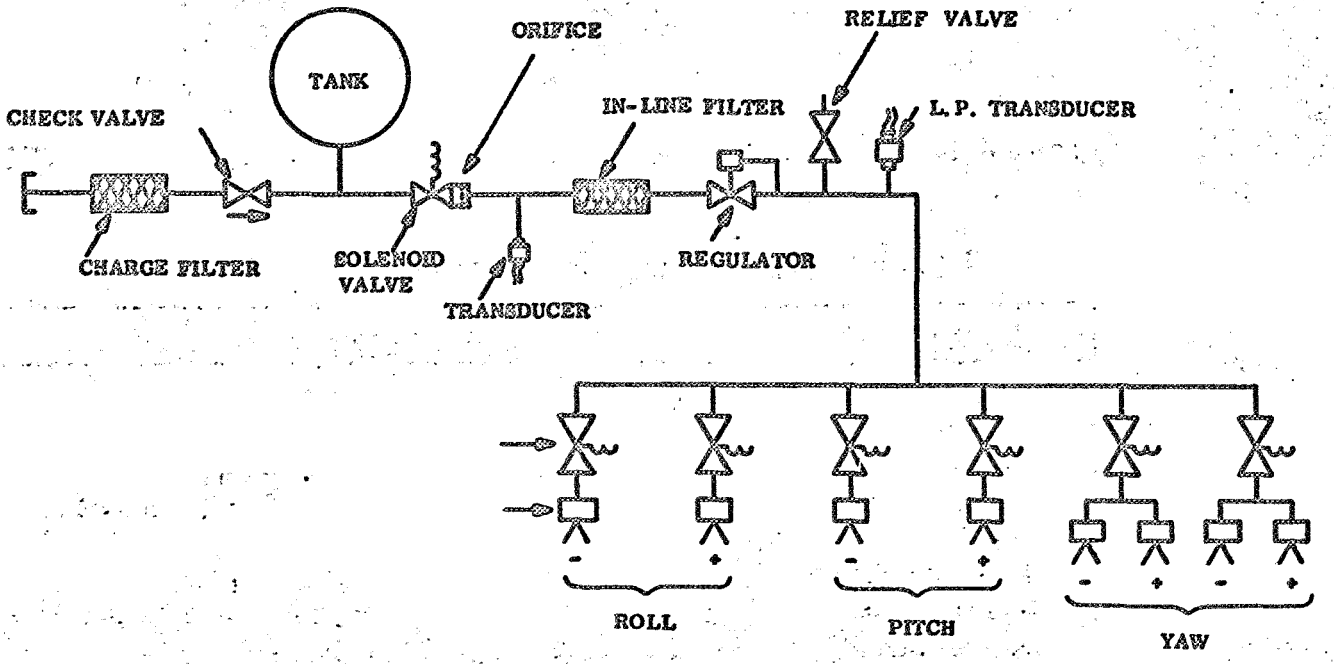


Figure 6-2. NIMCO Attitude Control System Schematic

SECTION 7

TEST PROCEDURES

7.1 FUNCATIONAL TEST PROCEDURES

The functional test procedures for the breadboard system are given in Appendix A. During each test, the system pressure, nozzle chamber pressure, regulated pressure, and system "on-time" were recorded on an oscillograph record. A sample tracing of the recorded output is given in Figure 7-1. The instrument was calibrated before each test to verify accuracy of results.

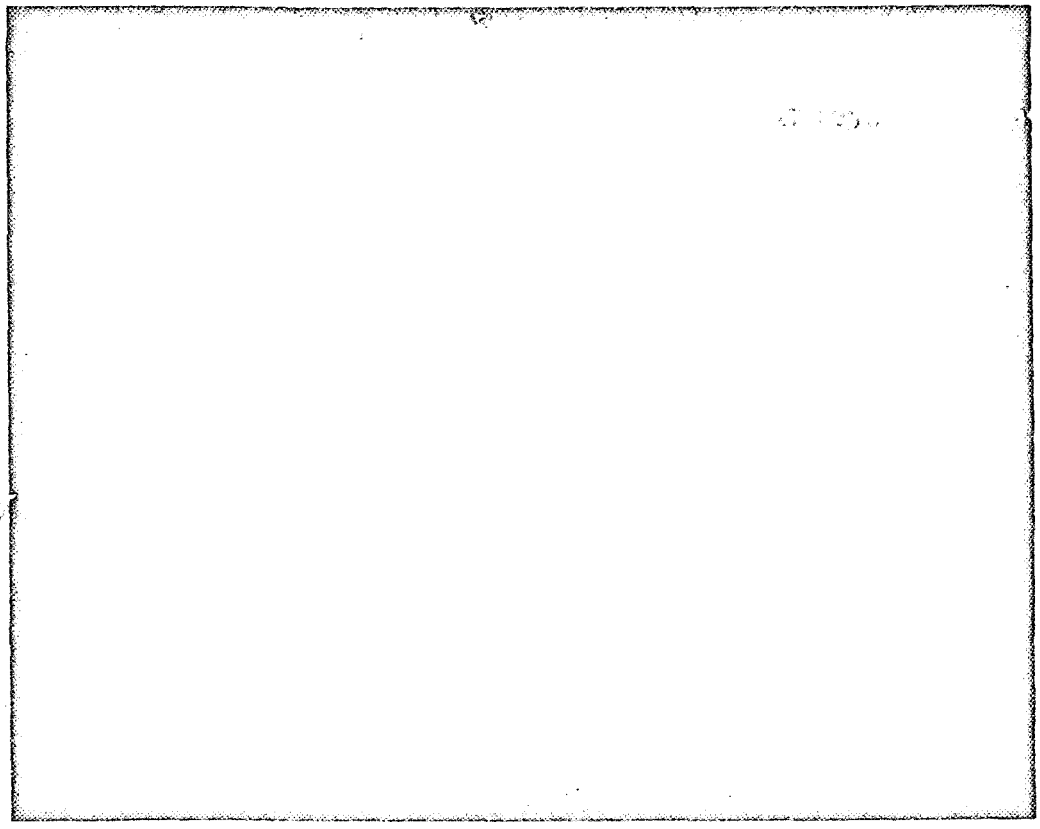


Figure 7-1. Oscillograph Trace

Prior to testing, the system was pressurized with nitrogen from a facility 5000-psi source located in the clean room. The facility gas was prefiltered through two 0.45-micron high-pressure filters* mounted in series near the inlet to the breadboard ACS. Vibration of the system was performed as part of the testing requirements for two of the four bread-

*Millipore Filter Corp., Bedford, Mass.

board system tests. Figure 7-2 illustrates the ACS on a vibration table. The fully pressurized system was vibrated in all three axes. The Voyager vibration frequency range and "g" loads were specified. However, in the system buildup, the pressurant tank selected was prime hardware from the Gravity Gradient Satellite (GGII) program and could not be vibrated higher than the GGII requirements. A vibration of 5 to 2000 cps at 3 g, 1 minute/octave was applied. All testing was conducted in a clean room to minimize the possibility of filter contamination during testing. Figure 7-3 shows the ACS test setup. Care was taken not to contaminate the filters in preparation for attachment to the ACS by using sterile forceps and petri dishes in the removal, storage and replacement of the cotton plugs from the filter holder inlet and outlet ports.

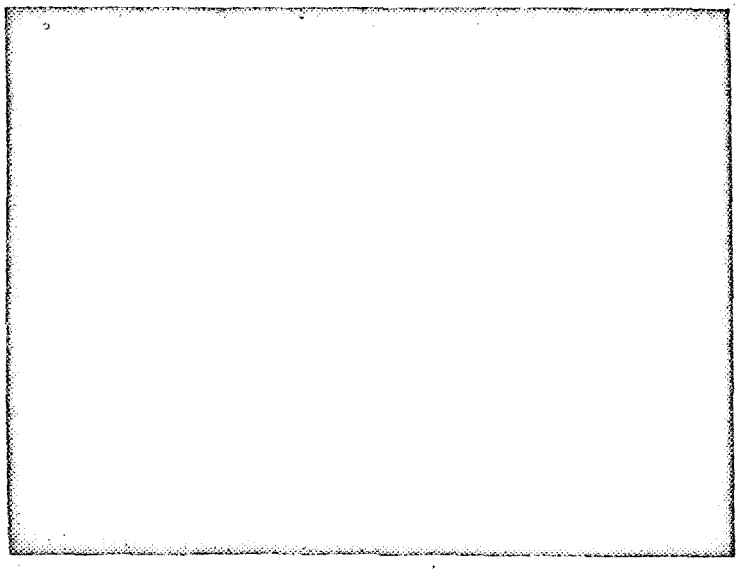


Figure 7-2. ACS on Vibration Table

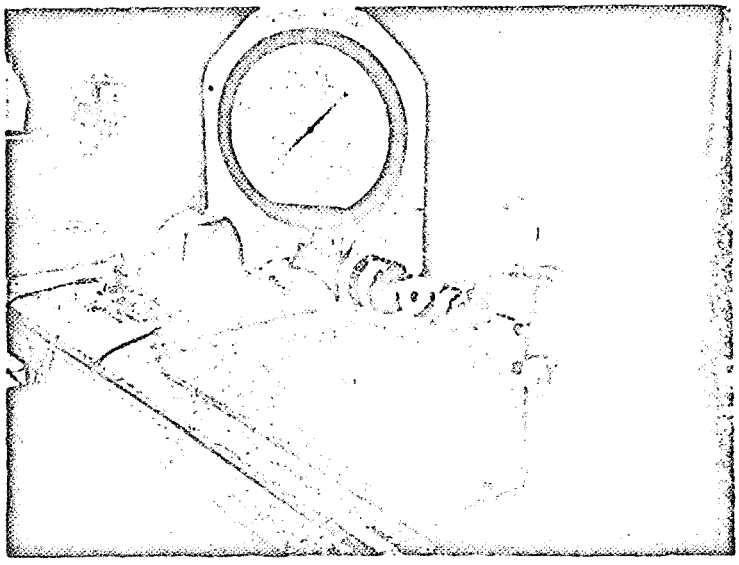


Figure 7-3. ACS Test Setup

7.2 CLEANING AND ASSEMBLY PROCEDURES

All the components were cleaned and assembled to meet the requirements of GE specification No. SVS 2631E para. 3.7, and Quality Control, Test Standing Instruction No. SI-236869 and the work statement. The applicable portions of the specifications are given in Appendix C. Assembly work was performed in a clean room designed to meet class 100,000 requirements. The actual room cleanliness level is indicated in the sample data sheet (Figure 7-4). Microbological contamination of the clean room was also monitored during test 6. Test data is given in Table 8-12. Specification SVS 2631E only specifies the particulate cleanliness level of the purge effluent from parts and the assembly. The parts degreasing (as required by the specification) was accomplished by ultrasonic cleaning in a freon bath those components that could be disassembled without affecting their performance. Components such as the regulator, transducer, and solenoid valves were cleaned by freon flush in the flow region and then purged to the particulate count requirement of the specification. The Quality Control logs showing particular count for each component and the system are given in Appendix E.

AREA 114209 Pneumatics lab. M & P NO.

CONTAMINATION CONTROL LABORATORY

Space Materials Research & Development

PARTICLE COUNT DATA

DATE SAMPLED 2/3/67 PARTICLE SIZE RANGE >1μ

DATE COUNTED 2/6/67 OPERATOR B. Lindley NOTEBOOK REF. Blue #252

ELECTROLYTE 1% H2SO4 in DMF 1 1 2 GAIN 64# APERTURE DIA. 100μ

SAMPLE NO.	AIR LIMIT 20,000 PARTICLES 1μ/cu. ft.	FLOOR LIMIT 150μ(270 parts.) 450μ(70 parts.)
Area #1 Room 1	10,000/ft³	
Room 2	11,200/ft³	
Area #2 Room 1	11,000/ft³	
Room 2	11,200/ft³	
Area #3 Room 1	6,700/ft³	
Area #4 Room 1	11,200/ft³	
Area #5 Room 1	2,650/ft³	
Anteroom	12,400/ft³	
Avg. Rm #2	11,550	
Avg. Rm. #1	7,910	

REMARKS:

APPROVED BY: E. C. Jones

DATE: 2/9/67

Figure 7-4. Contamination Control Laboratory Data Sheet

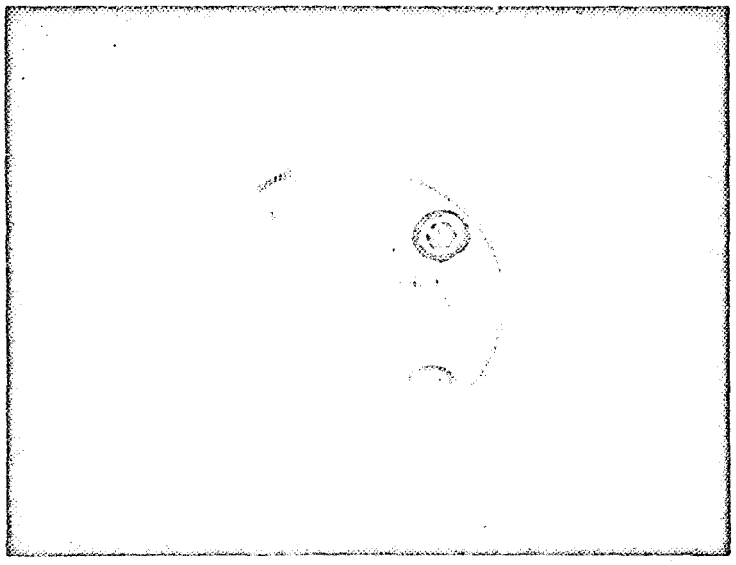
7.3 BIOASSAY PROCEDURES

The bioassay procedures are presented in detail in Appendix D. Theses procedures are adaptations of standard procedures recommended by NASA, U.S. Public Health Service, and standard manuals of microbiological techniques. The use of filters to sample airborne microbial contamination is described in U.S. Public Health Service Monograph No. 60, "Sampling Microbiological Aerosols." This filtration technique was adapted to measure contamination in the gas discharge of the attitude control system. High-pressure filter holders* and membrane filters were used for this purpose. Figure 7-5 illustrates the high-pressure filter and filter holder. Specifications for heat-shock procedures, ultrasonication, peptone water and agar media were taken from Standard Procedures for Microbiological Examination of Space Hardware, National Aeronautics and Space Administration, 1966. The plate count technique is a standard one found in the following references:

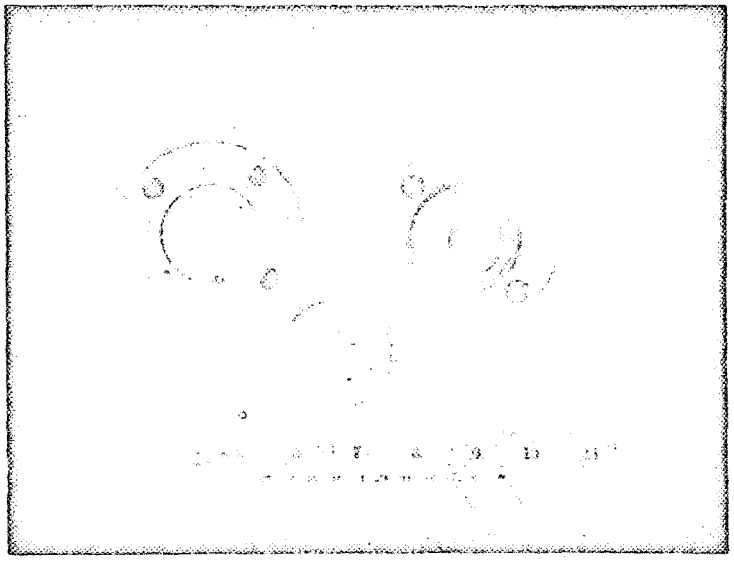
American Public Health Association, Inc.
Standard Methods for the Examination of Dairy Products, 1960

Society of American Bacteriologists
Manual of Microbiological Methods, 1957

*Gelman Instrument Co., Ann Arbor, Mich.



a. Gelman Filter Holder



b. Filter Holder and Filter Membrane

Figure 7-5. High-Pressure Filter and Filter Holder

SECTION 8

TEST RESULTS

Nine tests were conducted in this investigation. They are as listed below.

a. Four programmed tests with the breadboard system.

b. A single test of the NIMCO system.

c. One controlled contamination test with the breadboard system.

d. Three tests with the breadboard system to check system function and evaluate different bioassay techniques.

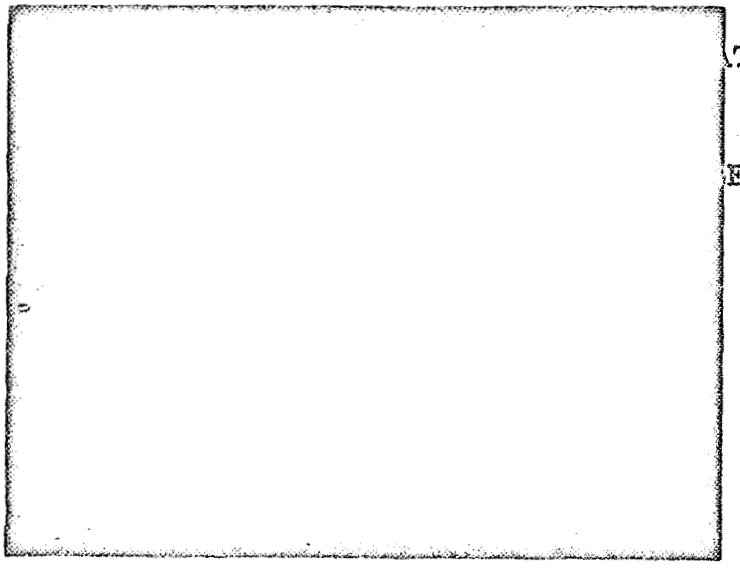
For the purposes of evaluating the ACS ejecta, only data from the test described in a, b, and c will be used.

All testing, cleaning, and bioassaying was monitored by members of the GE Quality Control Group for strict compliance to specification and procedures. Examples of QC logs are given in Appendix E. Prior to testing the breadboard ACS, a test was conducted with an auxiliary system (Figure 8-1). These tests were conducted to establish a standard bioassay technique to develop a method for installation of the ejecta filter, and to determine the pressure drop of the ejecta filters to assure that sufficient ΔP exists across the thruster to maintain sonic flow. Sonic flow is maintained when $\frac{P_a}{P_c} =< 0.528$* for nitrogen. With a thruster chamber pressure (P_c) of 51 psia, the nozzle discharge pressures (P_a) must not exceed 27 psia or 12 psia to maintain a $\frac{P_a}{P_c} < 0.528$. Table 8-1 shows the nozzle discharge pressure with filter membranes of 0.8 and 0.45 microns pore size at applicable flow rates. The thruster flow rate is approximately 0.0243 ft^3/min.

---

*Critical pressure ratio $\frac{P_a}{P_c} \left(\frac{2}{k+1}\right)^{\frac{k}{k-1}}$

k = ratio of specific heats of the gas. For nitrogen, k = 1.4.



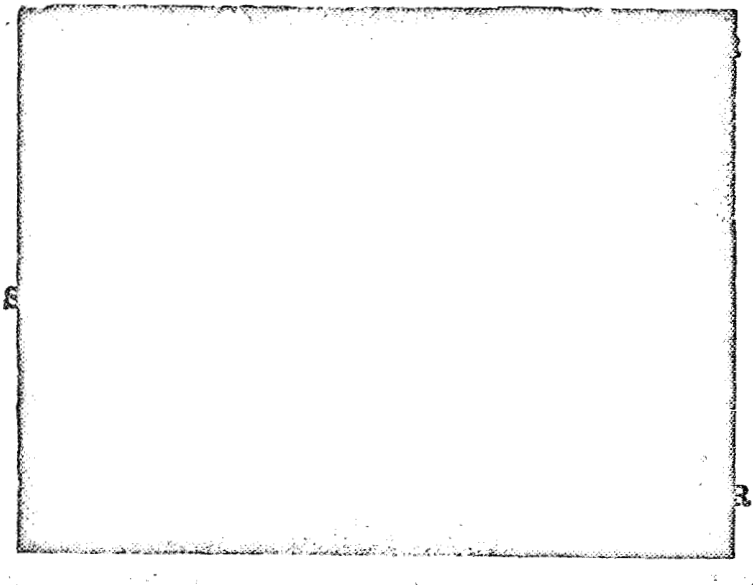


Figure 8-1. Auxiliary Test System

Table 8-1. Filter Tests

0.8-micron Pore Size Membrane Filter		0.45-micron Pore Size Membrane Filter	
Flow Rate	P_a	Flow Rate	P_a
1.0 scfm	0.6 psig	1.0 scfm	1.6 psig
1.4 scfm	0.8 psig	1.4 scfm	3.3 psig
1.8 scfm	1.5 psig	1.8 scfm	6.4 psig

The results of the tests show that using a 0.45-micron pore size filter element produces a $P_a < 12$ psig and does not impede nozzle flow.

The bioassay data presented in Tables 8-2 through 8-7 is broken down into two columns, the number of bacteria (including fungi) under the "nonheat-shocked" column, and the number of microbial spores under the "heat-shocked" column. Microbial recovery data given in Tables 8-2 and 8-3 are results obtained by bioassay procedure I (see Appendix D) and present the number of recovered organisms in the form "less than" (< X), because of the dilution factor in the process. Results given in Tables 8-4, 8-5, 8-6, and 8-7 are obtained by using bioassay procedure II in Appendix D and are actual bacterial colony counts.

The "control filters" are filters that were not exposed to the ejecta test but were bioassayed as controls with the ejecta contaminated filters. The assay of these filters provides the background contamination level of the bioassay procedure and also verifies the sterility of all the filters, since all the filters are sterilized together in a steam autoclave.

8.1 TEST 1

The first test followed Test Plan 1 (see Appendix A); that is, it had 21 filtered operations. During the testing, a leak was found between the nozzle and nozzle extension block after Step 14. The leak was fixed and the test continued. The bioassay data is given in Table 8-2. The leak was minute and did not leak sufficient gas to void the test. The data shows that no viable spores were recovered from 13 of the 18 filters assayed. Three filters were inadvertently contaminated. Of the five remaining filter elements, only one (Filter No. 9) had evidence of recovering any microorganisms from the ACS system. (Filter elements

1, 5, 6, and 11 (Step 7) provided inconsistent results; i.e., greater spore contamination was observed after heat-shock than the total count in the nonheat-shock, which may indicate contamination during the bioassay procedure.)

In general, the number of organisms recovered were so low that cultural techniques normally applied to enumerate the microbial population may not be sensitive enough. Because the biological laboratory background contamination obscures such low contamination levels, the test procedure was revised for Test 2 to allow more gas to flow through a filter for a particular system test. The revised test exposure (Test Plan II) allows two pulses to be passed through a filter before the filter is changed and also reduces the number of pulses in a test.

8.2 TEST 2

The breadboard system was subjected to a vibration of 5 to 2000 cps, 1 min/octave at 3-g level along all three axes. During vibration, the system pressure depreciated from 2000 psi to 1300 psi. Because of this loss of gas, only Steps 1 through 5 of Procedure II were followed, and the remaining gas was expelled through one filter. Table 8-3 provides the bioassay results of the test. Filter 1 was the only filter indicating viable spores. A total of 7.15 ft^3 of gas was bioassayed in this test.

8.3 TESTS 3 AND 4

In Test 3, the system was vibrated as in Test 2. Test 4 had no vibration. Test 3 and Test 4 were conducted to test Plan II. The bioassay procedure for these tests was changed. The new procedure placed 7 of the 15 filter elements directly on solid growth media (agar), and the remaining 8 were cultured as in Process 1 for heat-shock. All of the blended solution was cultured so as to recover a maximum bacterial population. This process would also enhance the detection of low numbers of contaminants. Results of these tests are given in Tables 8-4 and 8-5. Only three of the 29 filters assayed showed any indication of bacterial contamination.

8.4 TEST 5

A NIMCO attitude control pneumatic system which was installed in the engineering model satellite was tested for its ejecta bioloading. The NIMCO system was scheduled for testing as listed in Table 8-10. The bioassay testing was performed prior to the second scheduled thermal vacuum test (date 2/2/67). The NIMCO system was pressurized and functionally bench-checked as part of its planned test and had a remaining 700 psi in the propellant tank prior to the bioassay test. The test consisted of activating a positive roll and a negative pitch nozzle simultaneously for a 5-second pulse one for each nozzle. Six firings were made, using a total of 12 filters. Results of the bioassay are given in Table 8-6. The data shows a recovery of four viable organisms from two filters of the 12 used. A total of 1.46 ft^3 of gas was expelled.

8.5 OTHER TESTS

During the course of the above testing, it was necessary to run additional tests as checks on the system function and bioassay procedures.

A test was conducted to verify the quality of filtration of the facility-furnished filters. The facility series-redundant, 5000-psi line filters were removed, sterilized, and replaced in the system. The breadboard system was then pressurized with gas that was filtered by the facility filters, and all the gas was expelled through one discharge filter. All the filters used 0.45-membrane elements. The facility filters and the breadboard ACS discharge filter were bioassayed. The bioassay results are given in Table 8-7. No spores were cultured from any of the filters, and only 10 bacterial colonies were cultured from the first facility filter.

A controlled contamination test was performed as a final test, to determine what percentage of a viable test spore population would be exhausted from a system. A 0.1-ml solution of methyl alcohol with 1.13×10^8 spores of Bacillus subtilis var. niger was blown into the breadboard system, through a short tube, by the pressurinzing gas during the fill cycle. For this test the breadboard system in-line filter was removed. Steps 1 through 15 of Test Plan II (see Appendix A) were followed to complete the test. Results of the test are

given in Table 8-7. A total of 134 spores were recovered, of the test species type. A bioassay of the injection tube and system inlet check valve recovered 2.29×10^6 and 6.5×10^7 spores, respectively, or 62 percent of the injected test spores. These spores never entered the system. Higher recoveries may have been obtained had the injection process been more efficient. However, the recovery of some test organisms indicates that if contamination were present the recovery method could detect them.

8.6 BIOASSAY OF SYSTEM HARDWARE COMPONENTS

The internal surfaces of the breadboard ACS system in-line filter were bioassayed after the first test. The bioassay procedure for all hardware items is given in Appendix D. No viable organisms were found in the bioassay of this component. This test indicates that cleaning of the components, although primarily intended to reduce particulate contamination, provides a significant reduction in the level of microbiological contamination. To further assess the effectiveness of the system cleaning procedure on microbial contamination, four components (two in-line filters and two solenoid valves) of the same part number as that used in the breadboard system were bioassayed. One filter (Serial Number 33) and one solenoid valve (Serial Number 18) were cleaned according to the Nimbus specification (ultrasonic-cleaned in warm Freon Bath), and the other filter and solenoid valve purge-cleaned according to the Nimbus particulate requirements with no degreasing. The bioassay results are given in Table 8-9. Seventy-four spores were recovered from the filter, and none from the solenoid valve cleaned in the Freon ultrasonic bath. Cultures of two of the components cleaned by purging with gas provided spore recovery too numerous to count (TNTC).

Table 8-2. Test 1

Flow Time (sec.)	Gas Vol. (ft^3 @ 1 atm)	Filter Number	Average Number of Viable Organisms/ Membrane Filter	
			Nonheat-Shocked	Heat-Shocked
5.7	0.138	1	0	< 5
4.7	0.107	2	< 10	0
5.8	0.141	3	< 5	0
0.28	0.0067	4	0	0
0.3	0.0072	5	0	< 5
0.39	0.0093	6	0	< 5
180.00	4.36	7	< 10	0
5.8	0.140	8	< 30	0
5.6	0.134	9	< 5	< 5
5.9	0.142	10	< 10	0
0.42	0.01	11	0	0
0.32	0.0077	12	< 10	0
0.42	0.01	13	< 5	0
124.00	2.98	14	0	0
5.2	0.125	1	< 20	< 30, > 20
5.25	0.126	2	0	0
5.1	0.122	3	< 10	0
0.35	0.0084	4	< 30	0
0.26	0.0062	5	< 10	*
0.55	0.013	6	0	*
80.00	1.92	Blank	< 5	*
Fallout	0.367 organisms/petri dish-min or 0.00467 organisms/cm^2-min			

*Discarded because contaminated during one of the procedures.

Table 8-3. Test 2 (Post Vibration)

Flow Time (sec.)	Gas Vol. (ft^3 @ 1 atm)	Filter Number	Average Number of Viable Organisms/ Membrane Filter	
			Nonheat-Shocked	Heat Shocked
9.2	0.224	1	< 5	< 5
10.1	0.245	2	< 5	0
0.70	0.016	3	0	0
0.68	0.0155	4	0	0
180.00	4.36	5	< 10	0
109.00	2.65	6	< 20	0
Control		Blank	< 1	0

Table 8-4. Test 3 (Post Vibration)

Flow Time (sec.)	Gas Vol. (ft^3 @ 1 atm)	Filter Number	Average Number of Viable Organisms/ Membrane Filter	
			Nonheat-Shocked	Heat-Shocked
10.5	0.256	1	0	
0.73	0.0178	4	0	
182.0	4.44	5	0	
9.8	0.238	7	0	
0.67	0.0163	8	0	
11.1	0.220	11	0	
0.85	0.02	14	0	
55.0	1.34	Blank	0	
10.4	0.250	2		0
0.975	0.023	3		0
11.00	0.267	6		1
0.809	0.017	9		0
111.0	2.7	10		0
10.0	0.0243	12		0
1.2	0.029	13		0

Table 8-5. Test 4

Flow Time (sec.)	Gas Vol. (ft^3 @ 1 atm)	Filter Number	Average Number of Viable Organisms/ Membrane Filter	
			Nonheat-Shocked	Heat-Shocked
11.7	0.286	1	0	
11.0	0.269	2	0	
0.80	0.0195	3		1
0.76	0.0185	4		0
205.00	5.0	5		1
10.5	0.256	6		0
11.2	0.275	7	0	
0.66	0.016	8	0	
0.75	0.0183	9	0	
132.00	3.22	10	0	
11.9	0.29	11		0
10.1	0.25	12		0
1.08	0.026	13		0
122.0	2.74	14	0	
Control		Blank		0

Table 8-6. Test 5 (NIMCO)

Flow Time (sec.)	Gas Vol. (ft^3 @ 1 atm)	Filter Number	Average Number of Viable Organisms/ Membrane Filter	
			Nonheat-Shocked	Heat-Shocked
5.0	0.122	3	0	
5.0	0.122	4		0
5.0	0.122	5		3
5.0	0.122	6	0	
5.0	0.122	7	1	
5.0	0.122	8		0
5.0	0.122	10	0	
5.0	0.122	11		0
5.0	0.122	12		0
5.0	0.122	13	0	
5.0	0.122	14	0	
5.0	0.122	Blank		1
Control			0	3

Table 8-7. Test 6 (Control Contamination)

Flow Time (sec.)	Gas Vol. (ft^3 @ 1 atm)	Filter Number	Average Number of Viable Organisms/ Membrane Filter	
			Nonheat-Shocked	Heat-Shocked
11.3		1		0
10.2		2		0
1.11		3		0
1.20		4		17
184.00		5		33
11.5		6		0
10.8		7		17
0.89		8		0
0.90		9		0
123.00		10		50
10.75		11		17
12.2		12		0
0.95		13		0
116.0		14		0
Control		Blank		0

Number of test organisms (*Bacillus subtilis* var. *globigii*) introduced into system: 1.13×10^8

Number of organisms recovered from inlet tube: 2.28×10^6

Number of organisms recovered from check valve: 6.8×10^7

Table 8-8. Component Bioassay Test

Assay of Solenoid Valves and Filters	Total Number of Spores Recovered
Filter - Serial Number 3	TNTC
Filter - Serial Number 33	74
Valve - Serial Number 5	TNTC
Valve - Serial Number 18	0

Table 8-9. Clean Room Bioassay

Monitoring of Airborne Contamination of Clean Room Prior to Contaminated Test (Test 6)		
	Total Number of Organisms	
	Sedimentation Plates*	Reyniers Sample**
Day Before	60 (Assembly Room)	27 (Assembly Room)
	6 (Clean Room)	1 (Clean Room)
Day of Test	9 (Clean Room)	22 (Clean Room)
		11 (Clean Room)

The difference between levels of contamination in the clean room on the two days is probably due to the fact test personnel were working in the room on day of test but were absent with the exception of one person for twenty minutes on the day before.

*Exposed for 2 hours

**Sample of 60 cubic feet of air impacted against agar surface

Table 8-10. NIMCO Test Schedule

	Started	Completed
Open Bench Test	9-6-66	10-21-66
Assembly and Alignment	10-21-66	10-2-66
Functional Bench Test	11-3-66	11-9-66
Vibration	11-10-66	11-10-66
Post Vibration Alignment Check	11-11-66	11-14-66
Functional Bench Test	11-15-66	11-17-66
Thermal Vacuum Setup	11-17-66	12-5-66
Thermal Vacuum	12-5-66	12-9-66
Troubleshooting Repair and Retest	12-10-66	1-13-67
Open Bench Retest	1-19-67	1-20-67
Assembly and Alignment	1-20-67	1-25-67
Functional Bench Test	1-26-67	1-27-67
Vibration	1-30-67	1-30-67
Post Vibration Alignment Check	1-30-67	2-2-67
Functional Bench Test	2-2-67	2-4-67
Thermal Vacuum Setup	2-6-67	2-11-67
Thermal Vacuum	2-13-67	Not completed as of 2-16-67

SECTION 9

ANALYSIS

In general, the rate at which microorganisms are ejected from the attitude control systems tested was determined to be low. The quantity of organisms discharged is obscured by the normal background contamination level introduced during the bioassay procedure.

The background-introduced contamination was found at times to be at least equal to that expelled from the ACS at these low-contamination recovery levels. Because of the low contamination recovery levels, Test Plan II was introduced. This test plan increases the quantity of gas through each filter which would result in a lesser number of filters, higher concentration of contamination per filter, and a lower background contamination potential. Test II was performed using this procedure.

An alternate bioassay procedure was introduced (used in Test III and all following tests) for use when the ejecta contamination is expected to be very low. This procedure is described in detail in "Low-Level Contamination" in Appendix D. This procedure plates 7 of the 15 filter membranes from each single test directly on a growth medium, and the remaining filters are processed for spore detection. This procedure gives direct bacterial colony counts and further reduces the bioassay process contamination potential. The contaminant level of Test 3 and all remaining tests show only one bacterial colony and seven colonies from the heat-shocked aliquot from 41 filters bioassayed. The reduction of contamination recovered in these latter tests as compared with the earlier test is believed to be caused by the more efficient bioassay and test procedure, resulting in the reduction in bioassay procedural contamination. Tests 2, 3, and 4 would provide the most accurate ejecta contamination indications, since the control plates exhibit the presence of few or no contaminants. The NIMCO test correlates very well with the breadboard system test within the accuracy limits of the bioassay process.

The low biological discharge detected from the altitude control systems tested, and the low contamination level evident in the in-line bioassay would indicate a very clean system. The breadboard ACS, during its building, underwent a series of cleaning processes to

reduce primarily particulate contamination. However, the cleaning process also produces significant reduction in the level of microbiological contamination as evident from the component bioassay tests. The cleaning process responsible for the microbiological contamination reduction is the ultrasonic Freon degreasing process. This is shown by the results of the bioassay of the four components where two were ultrasonically cleaned in Freon and two were purged clean with gas. Not all the components in the breadboard ACS are cleaned in the Freon bath, and consequently, the total system is not at one level of biological cleanliness. In the breadboard system the regulator, transducers, and the solenoid valve were only purged-cleaned. It is further evident from this test that purging is a very ineffective means of removing biological contamination.

In the controlled contamination test, less than half of the injected spores enter the system beyond the system inlet check valve after being purged by the 10 ft^3 of gas required to fill the system. The injected spores were found plated on the walls of the inlet tube and check valve upon inspection prior to bioassay. The inability of the fill gas to remove the wet spores from the walls again shows that purging with gas does not remove substantial amounts of microorganisms from the surface of components.

SECTION 10

CONCLUSIONS

The results of this test program are consistent within the accuracy limits commensurate with biological evaluation of such systems, and it can be concluded that:

a. The level of biological contaminants ejected from an ACS that has been cleaned to the levels employed herein is extremely low. As a maximum, no greater than three viable colonies/ft^3 of gas were detected, and possibly, on the average, less than 1/ft^3 is ejected.

b. Based upon the sparce comparative data, the system size does not appear to significantly affect the microbial ejection rate.

c. Cleaning and assembly techniques within the current "state-of-the-art" can produce systems with low microbiological contamination ejecta potential.

d. The data obtained herein can be used to predict within reasonable accuracy the ejecta bioloading of the ACS Voyager system.

e. No conclusions can be drawn from this test concerning the effect of vibration and pulse mode operation on ejecta bioloading. However, because of the low ejecta quantity, vibration and pulse mode effects are not expected to be significant.

f. Purging a system with gas is a very ineffective means of removing organisms.

APPENDIX A

ACS SYSTEM TEST PROCEDURE

4.0	Procedure	
4.1	Clean all components to meet Spec. SVS 2631E - issued 6/5/65.	
4.2	Put system together according to drawing SK 36152-697.	
4.3	Purge system until system cleanliness meets Spec. SVS 2631E - issued 6/5/65.	
4.4	Install five galvanometers in CEC oscillograph recorder to pickup:	
4.4.1	Solenoid voltage	
4.4.2	Solenoid current	
4.4.3	Regulator inlet pressure (high-pressure transducer).	
4.4.4	Regulator outlet pressure (low-pressure transducer).	
4.4.5	Nozzle inlet pressure	
4.5	Installed seven galvanometers for reference lines.	
4.5.1	Nozzle inlet 0 PSI ref.	
4.5.2	Low-pressure transducer (greg. out)	30 PSI Ref.
4.5.3	Low-pressure transducer	35 PSI Ref.
4.5.4	Low-pressure transducer	41 PSI Ref.
4.5.5	High-pressure transducer (Reg. In)	0 PSI Ref.
4.5.6	Nozzle inlet	40 PSI Ref.
4.5.7	High-pressure transducer	500 PSI Ref.
4.5.8	High-pressure transducer	1000 PSI Ref.
4.6	Manufactured and installed protective circuits for galvanometers. Calibrated transducers to above references. After installing amplifier and transducer conditioner for nozzle inlet transducer.	
4.7	Run recorder to proper speeds during tests.	
4.8	Fill system with GN_2 to 2000 PSIG.	
4.9	Follow procedure 4.10.0 or 4.11.0, depending on which test plan is to be followed. Procedure 4.10.0 is for Test Plan I while procedure 4.11.0 is for Test Plan II.	

4.10 Test Plan I

4.10.1 Connect a sterile filter to ACS.

4.10.2 Fire one 5-second duration pulse.

4.10.3 Change filter to another sterile filter

4.10.4 Repeat step 4.10.2.

4.10.5 Repeat step 4.10.3.

4.10.6 Repeat step 4.10.2.

4.10.7 Repeat step 4.10.3.

4.10.8 Fire one 300-millisecond duration pulse.

4.10.9 Repeat step 4.10.3.

4.10.10 Repeat step 4.10.8.

4.10.11 Repeat step 4.10.3.

4.10.12 Repeat step 4.10.8.

4.10.13 Repeat step 4.10.3.

4.10.14 System blowdown to 50 percent pressure (1000 PSIG)

4.10.15 Repeat step 4.10.3.

4.10.16 Repeat steps 4.10.2 - 4.10.13.

4.10.17 System blowdown to 25 percent pressure (500 PSIG)

4.10.18 Repeat step 4.10.3.

4.10.19 Repeat steps 4.10.2 - 4.10.13.

4.10.20 System blowdown to 0 percent pressure (0 PSIG)

Test Plan II

4.11.1 Connect a sterile filter.

4.11.2 Fire two 5-second duration pulses.

4.11.3 Change filter to another sterile filter.

4.11.4 Repeat step 4.11.2.

4.11.5 Repeat step 4.11.3.

4.11.6 Fire three 300-millisecond duration pulses.

4.11.7 Repeat step 4.11.3.

4.11.8 Repeat step 4.11.6.

4.11.9 Repeat step 4.11.3.

4.11.10 System blowdown to 50 percent pressure (1000 PSIG)

4.11.11 Repeat step 4.11.3.

4.11.12 Repeat steps 4.11.2 - 4.11.9.

4.11.13 System blowdown to 25 percent pressure (500 PSIG)

4.11.14 Repeat step 4.11.3.

4.11.15 Repeat steps 4.11.2 through 4.11.9.

4.11.16 System blowdown to 0 percent pressure (0 PSIG)

APPENDIX B

Work Statement for Testing of a Cold Gas Attitude Control System for Ejecta Biossay

Prepared By: R. T. McFall 11-30-66
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Reaction and Actuation
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Approved By: C. C. Rich 11-30-66
C. C. Rich, Manager
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Test Plan

1.0 Test Objectives:

The objective of the test is to perform a complete test on a cold gas attitude control system. The test will define the amount of viable spores ejected from a cold gas ACS that has been cleaned and assembled under standard prescribed cleaning processes. The test will also show the effect of vibration on the spore loading of the ejecta.

2.0 Test Procedure:

2.1 Hardware Definition

2.1.1 Propellant tank - An Airite titanium nitrogen tank will be used. Vol. = 145 in^3, weight = 2.42 lbs, diameter = 6.730 inches and has a design pressure = 3600 psi.

2.1.2 Low pressure transducer - This is a Nimbus pressure transducer located on the low pressure side of the regulator to verify regulator performance.

2.1.3 High Pressure transducer - This advent pressure transducer shall be located on the high pressure side of the system to measure the internal gas pressure.

2.1.4 Fill and Vent Valve - The vent component is a Nimbus check valve located in the filling line to retain gas in the system upon removal of the ground charging line and filter.

2.1.5 In-line filter - This filter is an engineering OAO component. It is located in the system just before the regulator. It is rated for 10 microns absolute

2.1.6 Pressure Regulator - The pressure regulator is a gage type regulator (used on Nimbus) which regulates downstream pressure to 35 psi.

2.1.7 Solenoid Valve - The solenoid valve controls the flow of gas to the nozzles and shall be located adjacent to the nozzle. This valve is just like one used on Nimbus.

2.1.8 Discharge Orifice - These are Nimbus pitch axis nozzles which provide .099 ± 10% pounds thrust at steady state.

2.2 Hardware Preparation

2.2.1 Component

All components will be dissassembled in the component flow region, inspected and cleaned.

2.2.1.1 All parts which comprise the final assembly shall be critically examined to assure freedom from purns, feather edges, attached slivers, or any other similar type particles, which could subsequently become disloged.. Also check for cut or damaged seats, seals, etc. The examination shall also include a careful check of platings, coatings, or other finishes to assure that a satisfactory bond exists and that there is no danger of flaking, peeling, or other possible surface deterioration which could constitute a contamination hazard.

2.2.1.2 Cleaning shall be done in accordance with specification SVS 2631E dated 6-2-65, paragraph 3.7 in a freon bath with ultrasonic vibration. After each piece is reassembled and cleaned it should be purged and a milli pore count taken. The flush fluid, before entering the component shall be filtered to a 5 micron absolute level. The parts should meet the same level of cleanliness as the total assembly (2.2.2). Purge

components until the cleanliness level stated above is accomplished. Once cleaned, the components shall be bagged in cleaned plastic bags until assembly time, to maintain cleanliness.

A functional test should be performed on each component after cleaning and reassembly using filtered test fluids in such a manner as to not contaminate the component. (This requirement shall apply only to regulator, solenoid valve, and transducers).

2.2.2 System

System assembly techniques shall be in accordance with specification SVS 2631E dated 6-2-65, paragraph 3.7.4. Minimize the generation of particles. Precaution shall be taken to assure that the parts and sub-assemblies contain no residual cleaner, either as a liquid or as a solid residue. Handling of parts shall be accomplished only through the use of clean rubber gloves, tweezers, or other assembly tools, that have been cleaned properly.

All assembly work shall be done in a clean room. Assemble components on to vibration plate. After assembly is completed, the system must be purged until the final rinse solution contains no more than the number of particles shown below for 100 ml of solution filtered through a standard HA milli pore filter. The flush fluid, before entering the assembly shall be filtered to a 5 micron absolute level. Cleaning levels is from specification SVS 2631E dated 6-2-65, paragraph 3.7.4.

Particle Size	Maximum No. Particles	
	Metallic	Non-metallic (max. diam.)
16-25	No Requirement	
26	30	300
50	10	100

100	0	15
150	Non-allowable Maximum allowable fibers 50 x 500	

The specific cleanliness is only required on the internal surfaces of the system.

3.0 Calibration of Instrument

All instruments used must be recorded along with the calibration date, calibration period, and next calibration date. Any components that are calibrated (pressure transducer) must have complete data on the calibrating so it can be checked and verified.

4.0 Cold Gas - The propellant, gaseous nitrogen should be filtered to a .4 μ (Bio/inline) absolute level before entering the system and shall be dried to a dew. point of -65°F.

5.0 Testing

5.1 General

The cleanliness level of the assembly (Internal) shall be protected at all times during the testing operation and comply with specification SVS 2631E dated 6-2-65, paragraph 3.7.5 - 3.7.8. The inlet of the assembly shall be protected by external filters having a 5 - 10 micron absolute rating, cleaned in accordance with 2.2.1.2. If filters cannot be directly attached to the assembly, the intermediate fittings used shall be applied to the assembly in a clean room area, and shall not be removed until testing has been completed. Removal shall also take place under the same conditions. During testing, any other precautions necessary to assure that the assembly will remain clean shall be incorporated into the test procedure.

5.2 System Test

The system shall be loaded with biofiltered gas and tested as follows:

5.2.1 Initial Test

5.2.1.1 Three pulses of 5 second duration each. (step 1)

5.2.1.2 Three pulses of 30 ms duration each. (step 2)

5.2.1.3 System blowdown to 50% pressure.

5.2.1.4 Steps 1 and 2 repeated.

5.2.1.5 System blowdown to 25% pressure.

5.2.1.6 Steps 1 and 2 repeated.

5.2.2 Second Test

5.2.2.1 System vibrated to simulate boost phase.(1)

5.2.2.2 Three pulses of 5 seconds duration each. (step 2)

5.2.2.3 Three pulses of 30 ms duration each. (step 3)

5.2.2.4 System blowdown to 50% pressure.

5.2.2.5 Repeat steps 2 and 3.

5.2.2.6 System blowdown to 25% pressure.

5.2.2.7 Vibrate system to simulate retrofiring.(1)

5.2.2.8 Repeat steps 2 and 3.

5.2.3 Third Test

5.2.3.1 Repeat all steps in 5.2.1.

5.2.4 Fourth Test

5.2.4.1 Repeat all steps in 5.2.2.

5.2.5 Fifth Test

5.2.5.1 Biopsy of selected components (to be performed by MDL-Biosciences).

(1) Vibration rate comply with SVS 2631E issued 6-2-65, paragraph 3.4.2.2 (part B only)

Ejecta samples shall be taken for each firing of the system. After each test series the system is recontaminated by purging with compressed air, and reprocessed through the cleaning cycle before the next test. (2.2)

6.0 Documentation

The results of each test shall be reported and shall include data sheets prepared for the purpose. Copies of all original data sheets generated during the test program shall be included in this report. Each data sheet shall indicate the following:

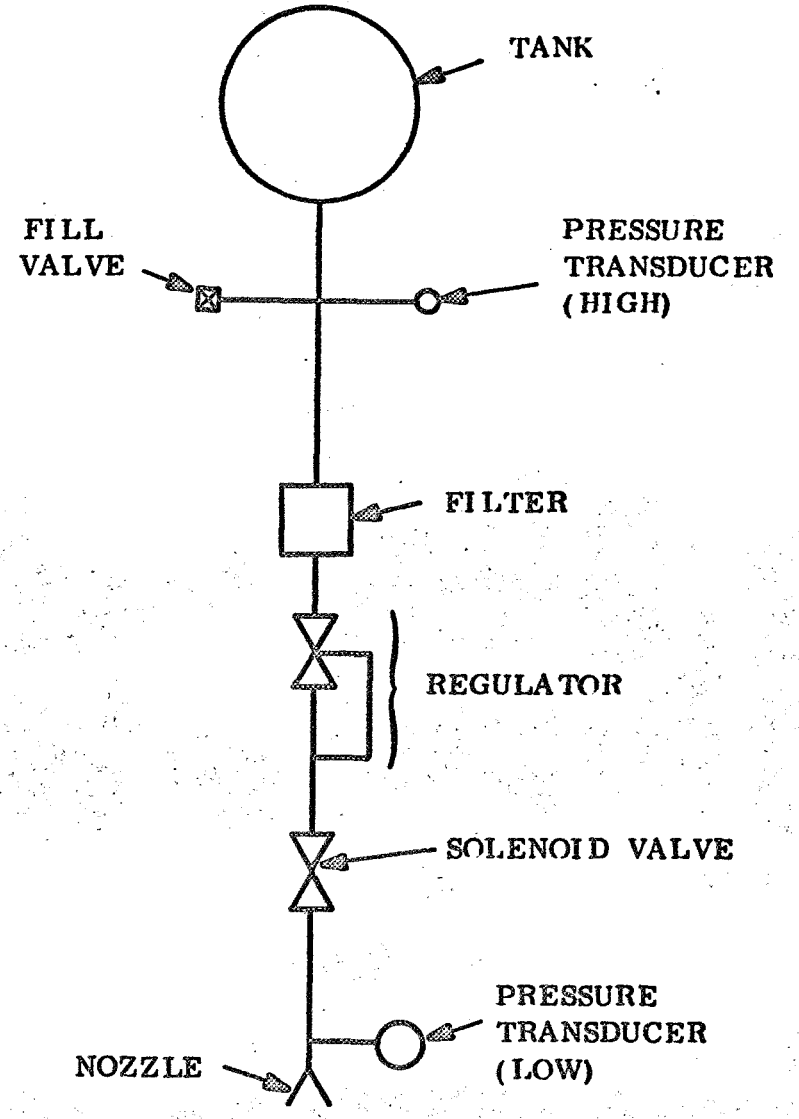
1. Name and drawing number and serial number of the component being tested.
2. Type test program being conducted.
3. Date of test.
4. Specifications and/or associated test procedure documents to which the Test is being conducted and the appropriate paragraph numbers.
5. Name and location of test facility.
6. Identification of test equipment employed including meters, gages, etc.
7. The specification limits for each value recorded.
8. Identification of each value recorded referenced to the specification, or associated test procedure document.
9. Clear identification of any and all "out-of-specification" readings.
10. Name of individual conducting test and names of any engineering witnesses including QC engineering witnesses.
11. Records to verify: Cleanliness of each component before each test, system cleanliness prior to each test, gas [illegible]tration during system charging, contamination purge after testing.
12. Name and location of the manufacturer of the component under test.
13. Time of beginning and ending each test.
14. Calibration date on all test instruments including calibration periods and next calibration date.

7.0 Recording of Data

All data shall be taken on a single oscillograph chart. Data shall include valve current, vibration levels, transducer outputs, and calibrations related to each.

8.0 Priority

Specifications SVS 2631E dated 6-2-65 governs this test plan and applies where applicable.



Single-Axis Attitude Control System

APPENDIX C

APPLICABLE PORTIONS OF SPECIFICATION: SVS 2631E AND SI 236, 869

SVS 2631E

3.7 Cleanliness - In order to meet the stringent leakage, life, environment, and reliability requirements of the system application, it is necessary that the utmost care be taken to assure that the assembly is free from actual or incipient contamination. Operations in Paragraphs 3.7.1 through 3.7.5 and 3.7.7 through 3.7.9 shall be accomplished in an area equivalent to a clean room. The procedures, methods, and operations listed below shall serve as a guide in achieving the cleanliness requirements delineated in Para. 3.7.4.

3.7.1 Part Degreasing - After all fabrication and test processes (except final assembly) have been completed, the parts which comprise the final assembly shall be thoroughly degreased. The parts shall be inspected under an ultraviolet lamp, 2500 to 3700 angstroms wave length, to verify that this condition has been attained. Thereafter, the parts shall be stored in plastic bags, or other containers, which are equally clean, until ready for assembly.

3.7.3 Inspection - Prior to assembly and after fabrication processes have been completed, all parts which comprise the final assembly shall be critically examined to assure freedom from burrs, feather edges, attached slivers, or any other similar type particles, which could subsequently become dislodged. The examination shall also include a careful check of platings, coatings, or other finishes to assure that a satisfactory bond exists and that there is no danger of flaking, peeling, or other possible surface deterioration which could constitute a contamination hazard.

3.7.4 Assembly Cleaning - Immediately prior to the assembly operation, all parts which constitute the final assembly (including shipping covers, port closures, shipping bags, etc.) shall be thoroughly cleaned using procedures and equipment which will provide an assembly which is clean to the level specified herein. The gases and liquids used in this process shall have been passed, finally, through a 5-micron absolute filter. The cleaning equipment shall be so arranged that contamination cannot get into the immediate area of the parts being cleaned. Assembly or other tools which will come in contact with the parts being assembled shall be cleaned to the same level as the parts. The parts shall be cleaned until the final rinse solution contains no more than the number of particles shown below per 100 ML of solution filtered through a standard HA millipore filter.

Particle Size (Microns)	Maximum Number Particles Metalic	Nonmetallic (max. dia)
16-25	No Requirement	
> 28	30	300
> 50	10	100
> 100	0	15
> 150	Nonallowable	
	Maximum allowable fibers:	50X500

3.7.5 Assembly - Assembly techniques which minimize generation of particles shall be used. Where the assembly is composed, or can be composed, of one or more sub-assemblies, these subassemblies shall be cleaned in accordance with Paragraph 3.7.4. Precautions shall be taken to assure that parts and subassemblies contain no residual cleaner, either as a liquid or as a solid residue. Handling of parts shall be accomplished only through the use of clean rubber gloves and tweezers, or other assembly tools, cleaned in accordance with 3.7.4 above.

3.7.6 Testing - The cleanliness level of the assembly shall be protected at all times during the testing operation. The inlet of the assembly shall be protected by external filters having a 10 micron absolute rating, cleaned in accordance with 3.7.4. If filters cannot be directly attached to the assembly, the intermediate fittings used shall be cleaned to the same cleanliness level as the assembly parts. If pressure measurements are required between any component and the external filters, the pressure tap shall also be provided with filters in accordance with the foregoing. The filters shall be applied to the assembly in an area equivalent to a clean room meeting area, and shall not be removed until testing has been completed. Removal shall also take place under the same conditions.

During testing, any other precautions necessary to assure that the assembly will remain clean shall be incorporated into the test procedure.

3.7.7 Exterior Cleaning - At the completion of testing, but prior to the removal of external filters, the assembly shall be cleaned externally using the methods and measurements of 3.7.4.

3.7.8 Final Cleaning - At the time the protective filters are removed from the assembly, final flushing and packaging shall be performed. Flushing shall be accomplished by flowing through the assembly with a liquid solvent or gaseous fluid which is compatible with the assembly materials and a flushing rate which shall be approved. The flushing time shall be sufficient to assure that all surfaces have been adequately exposed to the cleaning action. The flush fluid, before entering the assembly shall be filtered to a 5-micron absolute level. Flushing will continue until the particle count does not exceed that specified in 3.7.4.

3.7.9 Packaging - Precautions shall be taken to assure that the assembly contains no residual cleaner, either as a solid, liquid or gas. At the same time that the system is receiving its final cleaning, the packaging equipment shall be cleaned to the same or better cleanliness level.

The inlet charging port shall be capped in accordance to the requirements of Drawing 237R473, and the nozzle outlets capped with protective elastic rubber covers.

SI 236, 869

GENERAL ELECTRIC — STANDING INSTRUCTIONS — MISSILE AND SPACE DIVISION

SECURITY CLASS.	INITIALS	S.I. NO.
UNCLASSIFIED	DCS/vat	236,869

REVISION NO.	DATE	SUPERSEDES	SHEET NO.
1	10/9/63	9/12/63	5

(FOR USE OF GENERAL ELECTRIC EMPLOYEES ONLY).

3.0 CONTAMINATION

3.1 Airborne Contaminant -- It is required that the following airborne contamination limits be held for the semi-clean areas where the contaminant sensitive surfaces of the spacecraft must be exposed:

3.1.1 Operating Limit -- The following particle counts shall not be exceeded when made twice per week in the manner described in Para. 3.4.1 below:

Particle Size, microns	Particles/cubic foot
50 - 100	17
100 - 200	4
over 200	1

Records shall be kept on the airborne counts for all appropriate areas. No new area shall be used until two consecutive counts meets the above requirement. If routine counts in any one area exceed the values given, corrective measures shall be taken. These shall take the form of intensive cleaning, filter removal, etc., until operating norms are met. If the condition cannot be corrected in the same day, the incident shall be treated as a shutdown. (See Para. 3.1.2).

3.3.1 Pneumatics -- All work involving cleaning, assembly, installation, disassembly and repair of the pneumatics hardware shall be accomplished in existing clean room facilities. The standard operate and shut down condition for clean room area will apply.

It is mandatory that all of the pneumatic components be cleaned by purge with clean dry gas, prior to installation in the system. If these parts must leave the clean area before the system is sealed, they must be sealed in plastic for transport to one of the semi-clean assembly or test stations. (Ref. MSI 217389 & SVS 2631)

3.3.2 Slip Ring Assembly -- This item is in place in the vehicle when assembly level drilling takes place. It is a requirement that a dust tight cover be installed on this unit prior to any drilling and that the cover be cleaned prior to its removal after all such assembly level machine operations have been completed.

FORM 1016B REV. (10-62)

SECURITY CLASS. UNCLASSIFIED

APPENDIX D

TESTS FOR MICROBIOLOGICAL EVALUATION OF A COLD GAS ATTITUDE CONTROL SYSTEM

D.1 ENUMERATION OF THE VIABLE MICROORGANISMS EJECTED FROM A COLD GAS ATTITUDE CONTROL SYSTEM

D.1.1 INTRODUCTION

The release of microorganisms in the ejected gas from attitude control and thrust vector control motors has been indicated as a potential source of contamination of concern to the planetary quarantine control program. The tests set forth herein are designed to provide an evaluation of the levels of microbiological contaminants generated from such sources. The information obtained will provide information as to the control required, if any, to be imposed upon such systems.

The tests for enumerating the viable microorganisms ejected in the gas from such systems involve firing actual motors pressurized with typical gases into adequate chambers and recovering any viable microorganisms by filtering the exhaust gases and then performing a microbiological assay procedure upon the filter.

The following procedure has been prepared for the specific purposes of this experimental program and has employed, as much as possible, procedures or parts of procedures which have received some consideration or have been recommended as "Standards" for various groups for various purposes (References 1, 2, and 3).

D.1.2 PREPARATION AND STERILIZATION OF FILTER HOLDERS

1. Cognizant Propulsion Group personnel (Voyager Planetary Quarantine Study Study Task) shall load Gelman and/or Millipore high-pressure gas filter chambers with membrane filters of their choice. (This choice depends upon back pressure and flow rates involved.) At that time, they shall inform cognizant Biosciences Operation personnel as to which mem-

brane filters they employed, including brand, type, and pore size. Biosciences personnel shall log this information into Laboratory Log Book designated for these experiments.

2. Cognizant personnel of Biosciences Operation shall plug both holes of each chamber with nonabsorbent cotton. A large enough piece of cotton should be used so that there is good resistance when one attempts to pull it out after it has been screwed into place.

3. Biosciences Operation personnel shall place chambers in metal (aluminum mesh) baskets on their sides so that plugged ends stand free.

4. Sterilize chambers in baskets for 15 minutes at 15 psi of steam and 250°F using "Fast Exhaust" mode of autoclave.

5. Remove chambers in baskets immediately from autoclave at end of sterilization cycle. Staple sterilizer record into Laboratory Log Book.

6. Place baskets containing chambers in cabinet in Room M8619 designated "Sterile Hardware and Glassware Only." Discard and note any chambers on which sterility may have been compromised for any reason whatsoever (e.g., lost one of their cotton plugs). Sterilize and store chambers on their edges, not on the sides containing the cotton plugs.

7. Biosciences Operation will hold filter units in "bonded sterile storage." They will be delivered by Messrs. Kaplan, Bateson, and Koesterer or their designated representative to a cognizant individual from the Propulsion Group, Voyager Planetary Quarantine Study Task. Chambers will be logged into Laboratory Log Book when they are received from the cognizant individual or his representative, upon completion of experimental exposures. Chambers will be logged out of Laboratory Book when they are turned over to same.

D.1.3 MICROBIOLOGICAL ASSAY PROCEDURE

1. Log chambers back into Laboratory Book when they are turned over to or received from Propulsion Group cognizant individual. Log time of receipt, condition of chambers, and comments concerning exposure, compromised sterility, etc.

2. At opening of filter holder, visually inspect for integrity of filter in presence of Propulsion Group cognizant individual or Quality Assurance personnel. Note any irregularities in Laboratory Book.

3. Proceed with assay procedure as follows:

   (a) Remove membrane filter from filter holder by using sterile forceps on a bench in Room M8619, previously decontaminated with 2 percent Vesphene solution. Place filter in a sterile Waring blendor cup (semi micro monel) fitted with a cover. Add 50 milliliters of sterile 1 percent peptone water aseptically and macerate the filter membrane by blending at high speed for 3 minutes.

   (b) Blend or vortex all volumes or dilutions of fluid immediately before plating. If a blended filter has had to stand for longer than 1 minute before plating, place back on blendor and turn to high speed for a few seconds.

   (c) Plate three 5 -ml aliquots of original dilution (dilution factor = 1:10). Plate three 1-ml aliquots of original dilution (dilution factor = 1:50).

   (d) Heat-shock 25 ml of original dilution as follows:
   Aseptically pipette a 25.0-ml portion of the blended suspension into the bottom of a separate sterile test tube (200 x 25 mm), taking care not to contaminate the lip or the sides of the tube. The rinse fluid contained in each test tube is heat-shocked by placing in an 80°C water bath for 20 minutes. Make certain the water bath level is at least 1 inch above the level of the liquid contents of each test tube being heat-shocked. Tubes containing heat-shocked suspensions may be cooled by placing in a beaker of tap water immediately after completion of heat shock exposure.

(e) Plate three 5-ml aliquots of heat-shocked solution. Plate three 1-ml aliquots of heat-shocked solution.

(f) Plating procedures: Aseptically pipette aliquots indicated from blendor cup or heat-shocked test tube into 100-nm-diameter sterile petri plates (D.1.8). Add 20 ml of sterile, molten (45-50°C) Trypticase Soy Agar (D.1.9) to each plate and mix the contents by gentle swirling. Allow mixture to solidify.

(g) Pour one control plate for each batch of agar and one control plate for each package of disposable Petri dishes.

D.1.4 INCUBATION

All petri plates from above will be incubated in an inverted position aerobically at 32°C for at least 72 hours. The plates will be observed and any colonies forming will be counted after 24, 48, and 72 hours.

D.1.5 COUNTING PROCEDURE

Count colonies at 24 and 48 hours on Quebec Colony Counter with petri plate lid in place. At 72 hours, final count may be made with lid off. Count all colonies, including surface and sub-surface ones.

Use grids on Quebec Colony Counter as aid in counting. Start from upper left and count all colonies to right and above grid lines. Move in an orderly manner back and forth across plates so that colonies are neither missed nor counted twice. Record fragments of blended membrane filter from bacterial colonies. Differentiate count as follows: B = bacteria, F = fungi, and S = spreaders. The person performing assay and/or counting procedure shall be clearly identified as well as the date and time of each observation. Data record forms shall include untreated (raw data) as well as calculated results per filter. All processing and reduction of data shall be part of the data record sheet format.

D.1.6 CALCULATION AND REPORTING OF RESULTS

The average colony count from each of the three plates in each series (nonheat-shocked and heat-shocked, aerobic portions) multiplied by the reciprocal of the dilution factor equals the number of microorganisms and spores respectively entrapped on the filter. Report to Propulsion Group (Voyager Planetary Quarantine Study Task) the calculated average total number of organisms on the original filter from each filter holder. The average number of microorganisms or spores recovered divided by the volume of gas in cubic feet assayed will put the result on a unit basis for comparison and final reporting purposes. This last calculation, however, is the responsibility of the Propulsion Group. The data record of Biosciences Operation personnel shall be kept in bound paginated data books as currently employed by them for laboratory work. All entries shall be made in ink with no erasures. Alterations, changes, or additional notes and/or comments shall be added by crossing out the written material and making such alterations as required at that site. The filter holders shall be numbered individually and each run shall identify the specific ones employed.

D.1.7 CONTROLS

Early experiments should be performed simulating the entire assay procedure (for nonheat-shocked aerobic organisms at least) to verify the actual level of incidental contamination that may be obtained in the actual procedure. Presuming that the techniques will verify that only very low levels of incidental contamination result from the procedure, it would not be necessary to run one set each time, but only periodically throughout the period of experimental exposures.

D.1.8 HIGH-PRESSURE FILTER UNITS

Type

1. High Pressure Filter Holders:

   Gelman* Catalog Number 11101

   Millipore** Catalog Number XX45-047-00

* Gelman Instrument Company, Ann Arbor, Michigan
** Millipore Filter Corporation, Bedford, Massachusetts

2. Membrane Type Filters

Gelman* 47 mm GA (autoclaveable) type (or equivalent)

pore sizes 0.45 (GA-6) and 0.80 (GA-4) microns supplied in packages certified by the vendor as to the pore size, etc., of the filters.

D.1.9 PREPARATION AND STERILIZATION OF CULTURE MEDIA

D.1.9.1 1-Percent Peptone Water

Suspend 10.0 g of Bacto peptone** (or equivalent) in 1 liter of distilled water. Adjust to pH 7.0 with HCL or NaOH. Dispense as required (put approximately 52 ml into square screw capped bottles*** or standard dilution bottles). Sterilize by autoclaving at 121°C (250°F) for 15 minutes using the "liquids" cycle if an automated sterilizer is employed.

D.1.9.2 Trypticase Soy Agar****

Suspend 40.0 g dehydrated Trypticase Soy Agar (or equivalent) in 1 liter of distilled water. Heat gently with frequent agitation and boil for 1 minute. Dispense 300 to 350 ml into 500-ml screw-capped Erlenmeyer flasks or 600 to 650 ml into 1000-ml screw-capped flasks. Place caps loosely on flasks, and sterilize the 500-ml sized Erlenmeyer flasks by autoclaving at 121°C for 15 minutes and the liter size Erlenmeyer flask at 121°C for 20 minutes. If automated sterilizer is employed, select "liquid" cycle. After sterilization, remove flasks from sterilizer and allow to cool before tightening caps, or remove to 50°C water bath, if to be used immediately.

Sterilized agar in sealed, screw-capped flasks may be remelted no more than twice. Melting can be accomplished by placing in boiling water bath, or autoclave (just bring up to 15 pounds pressure for 3 minutes). All media should be stored in a clean closed cabinet or refrigerated, depending upon length of time to be held prior to use.

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*Gelman Instrument Company, Ann Arbor, Michigan
**Difco Laboratories, Incorporated, Detroit, Michigan
***Bussey Products Company, Chicago, Illinois
****Baltimore Biological Laboratories, Incorporated, Baltimore, Maryland

D.1.9.3 Petri Plates

Type

Commercially available, disposable, sterile 100-mm-diameter and 150-mm-diameter petri plates or glass petri plates of similar dimensions are acceptable.

Precaution

Sterile, disposable Petri plates have the advantage of being ready for immediate use, but occasionally the sterility of certain lots is jeopardized. Therefore, controls should be run from each separate package (sleeve) used. If reusable glass petri plates are employed, they should be washed thoroughly and resterilized. Specific procedures are available for this operation.

D.1.9.4 Cotton Swabs

Type

Commercially available cotton swabs (nonabsorbent cotton), firmly twisted to 3/16 inch by 3/4 inch long over one end of a 6-inch wooden applicator stick. The swabs shall be packaged individually into appropriate-sized test tubes, the tubes then being plugged with cotton or screw caps (loosened during sterilization) with swab heads away from the closure. Swabs can be sterilized by autoclaving at 121°C for 15 minutes.

D.1.9.5 Blender Apparatus

Semimicro monel blender cups* (or equivalent) with covers are satisfactory for macerating the membrane filter. They must be thoroughly cleaned in detergent solution, rinsed with distilled water and sterilized prior to use. Sterilize by autoclaving with lids ajar at 121°C for 15 minutes. If automated sterilizer is used, a "dry" cycle may be employed. The lids should be firmly in place when removing from autoclave and then allowed to cool in a clean area. Any blender base that has a slow and fast speed (8,000 and 15,000 rpm, respectively) is adequate for energizing the cutter assembly.

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*Available from Eberbach

D.1.9.6 Low-Level Contamination

When the number of microorganisms ejected is suspected or determined to be very low, it is advisable to resort to an alternate microbiological assay procedure in order to make sure that all organisms recovered are counted. A serious drawback to the procedure to be outlined below is that aggregates of bacteria may develop single colonies rather than multiple colonies. Because of this, the original procedure should be used to establish whether the level is less than 30 microorganisms on any one membrane filter. This being the case, the following protocol may be used.

All of the procedures described previously shall be employed except as follows:

a. Aseptically remove membrane filters from filter holders and place each filter on sterile solidified Trypticase Soy Agar in separate sterile Petri Dishes.

b. Pour just enough melted (less than 50°C) Trypticase Soy Agar over filters to cover them.

c. Allow agar to solidify, invert and place in 32°C incubator.

d. Count colonies that develop at 24 hours, 48 hours, 72 hours, and 1 week.

e. Record, calculate, report as outlined in Part I, Sections D.1.1 through D.1.6.

D.2 ENUMERATION OF VIABLE ORGANISMS ON THE INTERNAL SURFACES OF A COLD GAS ATTITUDE CONTROL SYSTEM

D.2.1 INTRODUCTION

As an adjunct to studying the control of microbiological contamination by means of assaying viable ejecta, it is useful to establish the level of surface contamination which can be tolerated on the hardware employed. Attitude control system hardware undergoes a series of cleaning procedures to reduce particulate contamination to a predefined level. Although only particulates are assayed, the cleaning procedures also produce significant reduction in the level of microbiological contamination. In addition, the firing of the attitude motors

itself may have an effect on the viability of microorganisms sloughing off hardware surfaces. Cleaning procedures and firing may thus be helpful in meeting Planetary Quarantine and Sterilization requirements. The procedure which follows has been generated in order to assay the internal surfaces of a cold gas attitude control motor system.

D.2.2 SECTION A - FLUSH TECHNIQUE

1. Flush a minimum of 100 ml of sterile Freon, 1 percent peptone solution or liquid culture medium through the component involved and collect in a sterile bottle. Use aseptic technique when opening the solution to be employed and when introducing it into the hardware component.

2. Plate three 5-ml aliquots of original dilution (dilution factor depends upon amount of rinse fluid used).

3. Plate three 1-ml aliquots of original dilution.

4. Aseptically pipette 25 ml of original dilution into a sterile test tube (200 x 25 mm) and and heat shock as outlined in D.1.3.

5. Plate three 5-ml aliquots of heat shocked portion.

6. Plate three 1-ml aliquots of heat shocked portion.

7. Incubate, count, record, calculate, report as outlined in Sections D.1.4 through D.1.6.

D.2.3 SECTION B - ULTRASONIC CLEANING

1. Plug ends of component which has received flush treatment with sterile rubber stoppers. If this is not possible, plug by some other means. If internal parts are to be ultrasonically cleaned, disassemble aseptically and place parts of interest (using sterile forceps) into a sterile bottle.

2. Decontaminante exterior of plugged component by exposing to Ethylene Oxide (D.2.4) for 4 hours or more in an Isolator (D.2.4) which contains sterile cotton swabs, 70 percent isopropanol, sterile forceps, other tools for opening component, and a bottle (opened) large enough to contain the component and its parts.

3. Exhaust ETO with room air for 1 hour or more using isolator fan.

4. Unplug component and, if nonsterile closures were used, swab ends with 70 percent isopropanol.

5. Disassemble component in isolator and place all parts in bottle set aside for this purpose. Close bottle.

6. Open isolator and remove bottle containing component parts

7. Aseptically add enough 1 percent solution to all bottles containing hardware of interest to cover all parts. Record amount of solution used in each case.

8. Treat bottles in Ultrasonic Bath (D.2.4) for 12 minutes. Make sure volume of liquid in bath is above that in bottle.

9. Plate three 5-ml aliquots of original dilution.

10. Plate three 1-ml aliquots of original dilution.

11. Aseptically pipette 25 ml or original dilution into test tube and heat-shock as before.

12. Plate three 5-ml aliquots of heat-shocked portion.

13. Plate three 1-ml aliquots of heat-shocked portion.

14. Incubate, record, count, calculate, report as outlined in Sections D.1.4 through D.1.6.

D.2.4 ULTRASONIC BATH

The ultrasonic bath (Branson Instruments: generator, A-300; tank, LT-80; power control, PC-30; or equivalent) shall conform to the following specification:

(1) The frequency shall be 25 kc/sec.

(2) The power output in relation to bottom surface area of the tank shall be at least 2.3 w/sq. inch (0.35 w/cm^2).

(3) If the ultrasonic bath is not automatically tuned, tuning shall be performed according to the manufacturer's directions.

(4) The inside surfaces of the bath shall be stainless-steel.

(5) Glass bottles containing piece-parts or stainless-steel strips shall be supported on the bottom of the tank.

(6) The tank fluid shall be an aqueous solution of 0.3 percent by volume polyoxyethylene sorbitan mono-oleate (Tween 80).

(7) The temperature of the bath fluid shall be at least 25°C and shall not exceed 37°C. The bath fluid shall be changed periodically in order to prevent heat-up.

(8) The bath liquid shall be at least 1 inch above the level of the liquid in the bottles being ultrasonicated.

D.2.5 PROCEDURE FOR SURFACE STERILIZATION EMPLOYING ETHYLENE OXIDE

D.2.5.1 Apparatus

The apparatus as normally used, consists of the following:

a. Cabinet

1. Flexible Isolator (Flexible plastic hood or exposure chamber of PVC)

(a) Port or opening for introduction of ETO

(b) Opening for loading isolator approximately 18" diameter with inner and outer covers

b. Decontaminating/Sterilizing Agent

The agent of choice is an ethylene oxide (ETO) dicholorodifluoromethane (Freon-12) mixture (12% ETO): 88 percent Freon-12 by weight) packaged in a liquified state in gas cylinders.

The proper introduction of the sterilant requires that the gas cylinders be attached, through pressure reduction, heat exchange and valving equipment, to the chamber.

D.2.5.2 Method of Operation

Insure that the air filtration system is in proper working condition and that every leak is repaired. Check the ETO heat exchanger and the gas supply tubing for leaks.

a. Remove the flexible plastic cover on the entrance port by removing the rubber band or "O-ring."

b. Place in the chamber the items to be tested or assayed. Also add any other equipment such as tools, sealed culture media, decontaminating agent that may be required. Make sure that no contact seals are made with any part of the chamber which might act as a barrier to permeation or penetration of the sterilant.

c. Seal the unit by replacing the cover on the opening and the rubber band, tape or O-ring.

d. Add the necessary hot water (60 to 80°C) to pail holding the heat exchanger (copper coil) leading from the ethylene oxide dispenser into the chamber.

e. Before charging the chamber with the sterilant, make sure that all of the ports of the filter apparatus are closed with rubber stoppers or plastic film coverings and that the isolator is collapsed as completely as possible.

Enter the sterilant as a gas. The gas (ETO) is slowly passed into the line by opening the valves from the chamber to the sterilant dispenser. First, open the gas cylinder valve; then slowly open the valve leading to the heat exchanger. When the hood is inflated and the predetermined amount of ethylene oxide has been added, turn off the valve regulating the ETO and let the hood decontaminate for the time required. (A minimum of 6 hours is recommended with an overnigh. period of 12 to 16 hours desirable.

f. After the decontamination treatment, aerate the chamber by passing air through one of the complete sterile air filtration apparati through the isolator and out a second filter. Breathing of the chemical in the surrounding area should be avoided.

g. Proceed with disassembly of component as outlined in Section D. 2. 3.

D. 2. 6 REFERENCES

1. American Public Health Association, Inc., 1960 Standard Methods for the Examination of Dairy Products.

2. Manual of Microbiological Methods 1957; Society of American Bacteriologists, McGraw-Hill Book Co., In., New York.

3. Standard Procedures for the Microbiological Examination of Space Hardware, June, 1966, National Aeronautics and Space Administration.

APPENDIX E

SAMPLES OF QUALITY CONTROL LOGS

PERFORMANCE HISTORY

NAME Cold Gas Attitude Control System	DRAWING NUMBER	A N	PROGRAM
E SERIAL NUMBER	TEST INSTRUCTION NUMBER 611 M3-017	REV.	DATE
ENVIRONMENT	TESTER PERFORMING M L BRIDE		
QUALITY CONTROL ENGINEER	TEST CONDUCTOR		

TIME	REMARKS

EQUIPMENT NAME	MAKE	MODEL NO.	IDENT. NO.	CALIBR. DUE
(1) TANK - PART # 6481-1				
SERIAL # 0001				
MFG. SARGENTO/AIRITE DIV.				
PROOF PRESSURE. HYDROSTATIC TESTED				
TO 3500 PSIG.				
CLEANED WITH FREON - AND DRIED WITH				
GN2.				

PARTICLE COUNT.

SIZE (MICRONS)	METAL	NON METAL
>26	8	45
>53	2	18
>100	0	6
>150	0	0

OPERATING TIME ON THIS PAGE	CYCLES	HRS.	MIN.	ACCUMULATED TIME TO DATE	CYCLES	HRS.	MIN.	INITIALS

PERFORMANCE HISTORY

NAME Cold Gas Attitude Control System	DRAWING NUMBER	A N	PROGRAM Ejectm Bypass
& SERIAL NUMBER	TEST INSTRUCTION NUMBER 41M3-017	REV.	DATE

ENVIRONMENT	TESTER PERFORMING MC BRIDE
QUALITY CONTROL ENGINEER	TEST CONDUCTOR

TIME | REMARKS

EQUIPMENT NAME	MAKE	MODEL NO.	IDENT. NO.	CALIBR. DUE
REGULATOR -	CARLETON - 1136 7254 P1 - S/N 1			
LOCK UP - 38.6 PSIG				
FLOW @ 300 PSIG INLET				
FLOW SCFM	OUTLET PSIG	LOCKUP PSIG 38.6		
1	38.3			
2	38.0			
3	37.7			
4	37.3			
5	36.5			
RELIEF VALVE TEST				
CRACK -	72.5 PSIG			
RESEAT -	64.0 PSIG			
SEAT LEAKAGE -	0 PSI IN 30 MIN			
PARTICLE COUNT -				
SIZE (MICRONS)	METAL	NON METAL		
>26	3	23		
>50	1	15		
>100	0	1		
>150	0	0		

OPERATING TIME ON THIS PAGE	CYCLES	HRS.	MIN.	ACCUMULATED TIME TO DATE	CYCLES	HRS.	MIN.	INITIALS

PERFORMANCE HISTORY

NAME LOX GAS ATTITUDE CONTROL SYSTEM	DRAWING NUMBER	A N	PROGRAM ELECTA BIOASSY
SERIAL NUMBER	TEST INSTRUCTION NUMBER 41113-017	REV.	DATE
ENVIRONMENT	TESTER PERFORMING MCBEIDE		
QUALITY CONTROL ENGINEER	TEST CONDUCTOR		

TIME	REMARKS

EQUIPMENT NAME	MAKE	MODEL NO.	IDENT. NO.	CALIBR. DUE
CHECK VALVE	CIRCLE SEAL	SK5610S-07E-6	S/N 102284	
FLOW AT 12 PSIG - .14 SCFM				
FLOW TEST				
INLET PSIG	OUTLET PSIG	FLOW SCFM		
100	80	6 SCFM		
LEAKAGE				
INLET PSIG				
200 -	.012 CC/MIN			
3250 -	.011 CC/MIN			
PARTICLE COUNT				
SIZE (MICRONS)	METAL	NON METAL		
> 26	1	0		
> 50	7	3		
> 100	0	0		
> 150	0	0		
EQUIPMENT USED				
GAUGE	HEISE	0-300	ON 0151	1-3-67
GAUGE	HEISE	0-100	ON 0033	1-13-67
FLOWMETER	F. SMITH/PORTER	GLASS/METAL	ON 0082	3-1-67
FLOWMETER	F. SMITH/PORTER	VALVE/METAL TUBE		6-1-67
LEAK DETECTOR	CEC	24-120	10-318	3-11-67

OPERATING TIME ON THIS PAGE	CYCLES	HRS.	MIN.	ACCUMULATED TIME TO DATE	CYCLES	HRS.	MIN.	INITIALS

PERFORMANCE HISTORY

PAGE 4

Control System	DRAWING NUMBER	A N	PROGRAM Biomass
E SERIAL NUMBER	TEST INSTRUCTION NUMBER 41M3-017	REV.	DATE
ENVIRONMENT		TESTER PERFORMING McBride	
QUALITY CONTROL ENGINEER		TEST CONDUCTOR	

TIME	REMARKS

EQUIPMENT NAME	MAKE	MODEL NO.	IDENT. NO.	CALIBR. DUE
Solenoid Valve	Valcor	V27200-53	S/N 11	
Leakage				
Inlet PSIG				
50 psig	.0112 cc/sec	Total Leakage		
Response Time				
Open	13 milliseconds			
Close	3 milliseconds			
Particle Count				
Size (microns)	Metal	Non Metal		
>25	1	5		
>50	0	1		
>100	0	0		
>150	0	0		
Equipment Used				
Leak Detector	CEC	24-120	UA 0121	3.11.67
Accelerometer	CEC	5-116	700316	

OPERATING TIME ON THIS PAGE	CYCLES	HRS.	MIN.	ACCUMULATED TIME TO DATE	CYCLES	HRS.	MIN.	INITIALS

E-4

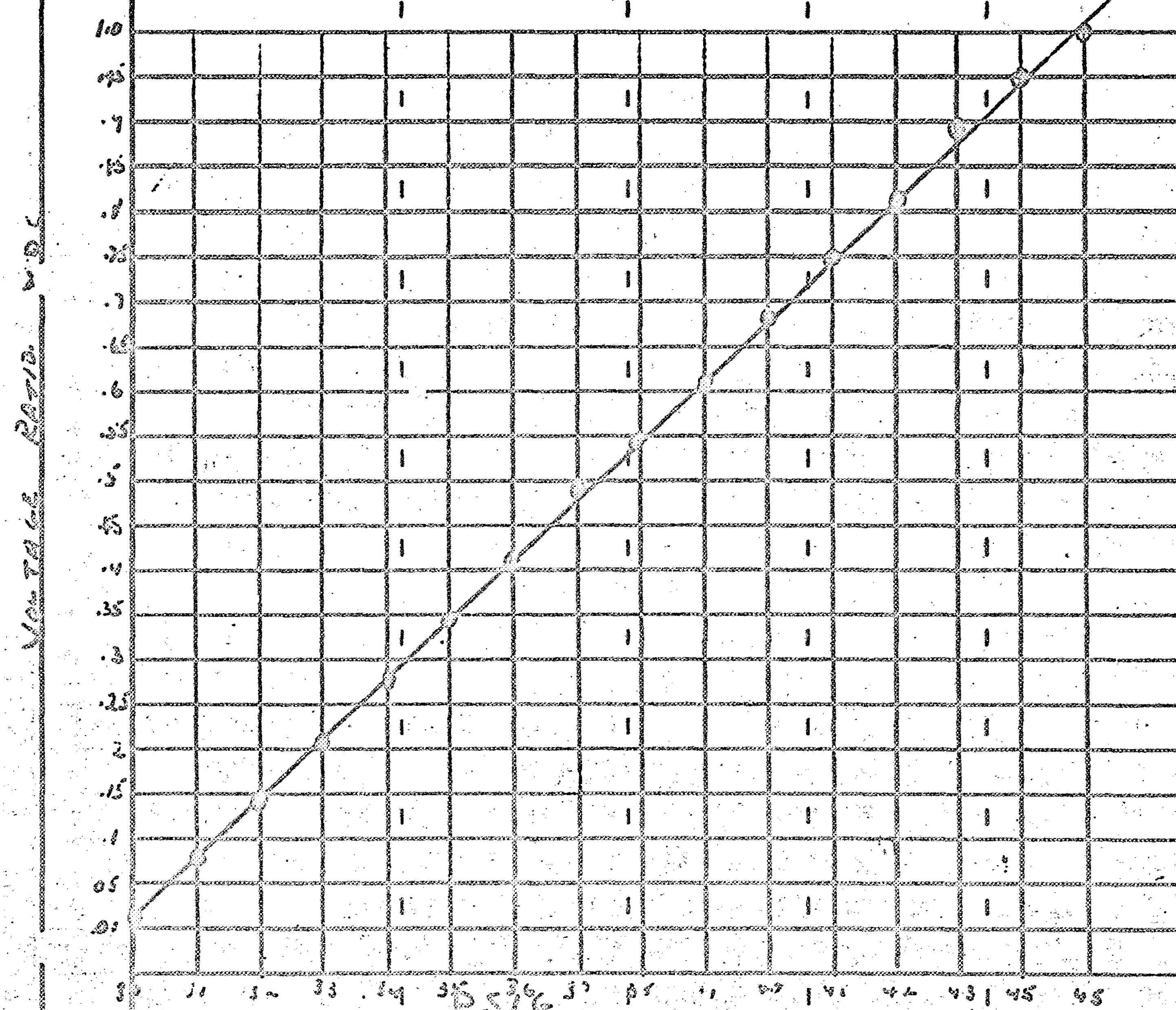
PERFORMANCE HISTORY

NAME COLD GAS ATTITUDE CONTROL SYSTEM	DRAWING NUMBER	A N	PROGRAM 2JE474 BIOSASSY
& SERIAL NUMBER	TEST INSTRUCTION NUMBER 41 M 3-017	REV.	DATE

ENVIRONMENT	TESTER PERFORMING McBRIDE
QUALITY CONTROL ENGINEER	TEST CONDUCTOR

TIME	REMARKS

EQUIPMENT NAME	MAKE	MODEL NO.	IDENT. NO.	CALIBR. DUE
LOW PRESSURE TRANSDUCER	(TRANSONIC)	1478590 5PL		S/N 89483
GAUGE	HEISE	0-300	34 3151	1-2-67
RATIOMETER CALIBRATION -	WALLACE TIERNAN	703300-20	-TD036	2-30-67



OPERATING TIME ON THIS PAGE	CYCLES	HRS.	MIN.	ACCUMULATED TIME TO DATE	CYCLES	HRS.	MIN.	INITIALS

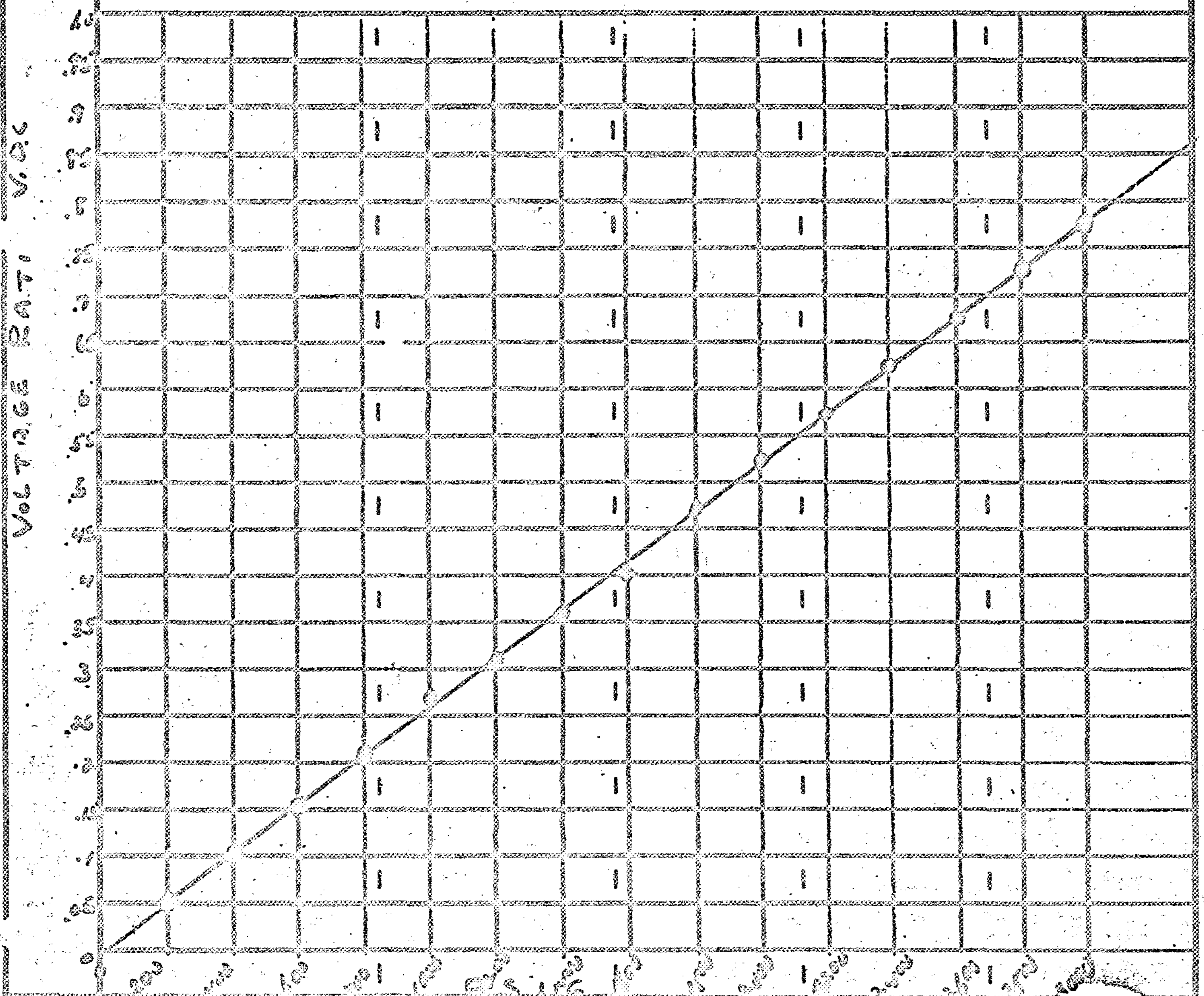
PERFORMANCE HISTORY

PAGE 6

NAME	DRAWING NUMBER	AN	PROGRAM
Cold Gas Attitude Control System.			[illegible]
SERIAL NUMBER	TEST INSTRUCTION NUMBER	REV.	DATE
	[illegible]		

ENVIRONMENT	TESTER PERFORMING
	[illegible]
QUALITY CONTROL ENGINEER	TEST CONDUCTOR

TIME	REMARKS			
	EQUIPMENT NAME	MAKE	MODEL NO.	IDENT. NO.	CALIBR. DUE
	High Pressure Transducer.	Transonic	2671	[illegible]	
	Gauge	Heise	0-5250	[illegible]	[illegible]
	Ratiometer	[illegible]		[illegible]	2-32-69
	Calibration				



OPERATING TIME ON THIS PAGE	CYCLES	HRS.	MIN.	ACCUMULATED TIME TO DATE	CYCLES	HRS.	MIN.

PERFORMANCE HISTORY

NAME COLD GAS ATTITUDE CONTROL SYSTEM	DRAWING NUMBER	A N	PROGRAM EJECM BIOASSY
& SERIAL NUMBER	TEST INSTRUCTION NUMBER 41M3-019	REV.	DATE

ENVIRONMENT	TESTER PERFORMING MCBRIDE
QUALITY CONTROL ENGINEER	TEST CONDUCTOR

TIME	REMARKS				
	EQUIPMENT NAME	MAKE	MODEL NO.	IDENT. NO.	CALIBR. DUE
	FILTER	A.P.M.	ACJ-3102-4190	S/N 7	
	PARTICLE COUNT				
	SIZE (MICRONS)	METAL		NON METAL	
	>26	1		2	
	>50	1		2	
	>100	0		4	
	>250	0		0	

OPERATING TIME THIS TEST	CYCLES	HRS.	MIN.	ACCUMULATED TIME TO DATE	CYCLES	HRS.	MIN.	INITIALS

PERFORMANCE HISTORY

NAME	DRAWING NUMBER	A N	PROGRAM
Pneu. Subsystem			ESE 670 Bosassy
S SERIAL NUMBER	TEST INSTRUCTION NUMBER	REV.	DATE
	4103-07		1-3-67

ENVIRONMENT	TESTER PERFORMING
	McBride
QUALITY CONTROL ENGINEER	TEST CONDUCTOR

TIME | **REMARKS**

EQUIPMENT NAME	MAKE	MODEL NO.	IDENT. NO.	CALIBR. DUE

SUBSYSTEM ASSEMBLED - PARTICLE COUNT TAKEN PER SVS 2631E PARA 3.7.4

PARTICLE SIZE (MICRONS)	METAL	NON METAL
>26	6	12
>50	3	10
>150	0	5
>350 (Per SVS 2631E)	0	0

INSTRUMENTATION USED -

NAME	MAKE	MODEL #	IDENT #	CAL. DUE
Millimicron	GES	S-144	TD-0[illegible]	1-28-67
Amplifier	Alinco	512A	TD-0117	
Power Conditioner	BRP	1-1137	TD-0031	9-28-67
Barometer	Leonard	700300-2	TD-0036	8-30-67
Power Supply	Universal	6-3501	OC0701	3-7-67
Gauge	Heise	H17532	ON0017	7-11-67

OPERATING TIME ON THIS PAGE	CYCLES	HRS.	MIN.	ACCUMULATED TIME TO DATE	CYCLES	HRS.	MIN.	INITIALS

Test No. 7 Filter No. 1-7

MISSILE AND SPACE DIVISION

QUALITY CONTROL PLANNING

Sheet 1 of 4

DWG. NO.	NAME: Ejecta - Bio Assay	PROJECT: Voyager
N.H.A.	APPLICABLE SPECS.	

OPN. NO.	DEFECT CLASS.	INSPECTION DESCRIPTION	TOOLS AND GAGES	INSP. STAMP
1		Cleanliness & Sterilization		
		Monitor Tools & Area		
		a) Sterile forceps ✓		
		b) Sterile petri dishes (12) per filter ✓		
		c) Sterile Waring Blender Cups (21) ✓		
		d) Sterile pipettes - 5.0 ml., 25.0 ml. ✓		
		e) Clean work areas. ✓		
2		Blending		
		a) Sterile forceps used to remove filter paper ✓		
		Filter in fact		
		b) Sterile Blender Cup used. ✓		
		c) 50 ml. sterile 1% peptone water added to Blender cup with filter-mouth of bottle passed over Buensen Burner. ✓		
		d) Mixture blended for 3 min - Mechanical Timer ✓		
		e) Log time of Blending 1634 1-3-67		

REV.					PLANNER	DATE
DATE & INIT.						1/3/67

FORM 1082V REV. (4-63)

E-13

MISSILE AND SPACE DIVISION

QUALITY CONTROL PLANNING

DWG. NO.	NAME: Ejecta-Bio Assay	PROJECT– Voyager
N.A.	APPLICABLE SPECS.	

OPN. NO.	DEFECT CLASS.	INSPECTION DESCRIPTION	TOOLS AND GAGES	INSP. STAMP
3		Assay		
		The following steps must be taken no later than 2 hrs.		GEASY-22
		after Blending - Log Time. 1537 1-3-67		
		Non-Heat Shocked Portion		
	ABC.	3ea. AND 3EA 1.0ml Portions DEF. a) 5.0 ml portions of Blender Cup aseptically pipetted into 100 ml sterile petri dishes. Pipette passed over flame.	✓	GEASY-22
		b) 20 ml (approx) of sterile (50) trypticase VIII soy agar added to petri dish-Pipette passed over flame.	✓	GEASY-22
		c) Mixture gently swirled and mixed - Log time & date completion. 1410 1-5-67		GEASY-22
		d) Mixture allowed to solidify.	✓	GEASY-22
		Heat Shocked Portion		
		5) 25.0 ml portion from Blender cup aseptically pipetted into bottom of sterile test tube, sides of test tube not contaminated by mixture. Test tube & pipette passed over flame.	✓	GEASY-22
		b) Test tube heat shocked in a 80-85°C water bath for 20 min. Mechanically timed. Insure water level a min of 1" above contents of tube.	✓	GEASY-22

REV.					PLANNER	DATE
DATE & INIT.					H Nielsen	1-3-67

Test No. Filter No 1-7

MISSILE AND SPACE DIVISION

QUALITY CONTROL PLANNING Sheet 3 of 4

DWG. NO.	NAME Ejecta-Bio Assay	PROJECT-- Voyager
M.A.	APPLICABLE SPECS.	

OPN. NO.	DEFECT CLASS.	INSPECTION DESCRIPTION	TOOLS AND GAGES	INSP. STAMP
	G.M.I	c) 5.0 ml. portions of heat shocked liquid (3-ea. AND 3-ea 1.0 ml Portions) aseptically pipetted into each of (6) 100 ml. dia. petri dishes (sterile). Pipette passed over flame.		
		d) Approx. 20 ml. of sterile, molten (50C) Trypticase Soy Agar added to each dish. Pipette passed over flame.		
		e) Contents mixed by swirling gently 10 times in each direction.		
		f) Log completion time & date. 16:40 1-3-67		
		g) Mixture allowed to solidify.		
4		Incubation		
		a) All (6) petri dishes placed in an inverted position in an over held @ 32°C for 72 hrs. - Log starting time and date. 12:00 1-3-67		
		b) Monitor oven temp. once every 8 hrs.		
		0 Hrs. - 32°		
		8 Hrs. - 31°		
		16 Hrs. - 32° -0730		
		24 Hrs. - 31°		
		32 Hrs. - 31°		
		40 Hrs. - 31°		

REV.					PLANNER	DATE
DATE & INIT.						1-3-67

FORM 10624V REV. (4-63)

E-15

Test No. Filter No. 1-7

MISSILE AND SPACE DIVISION

QUALITY CONTROL PLANNING

DWG. NO.	NAME	PROJECT
I.A.	Ejecta-Bio Assay	Voyager
	APPLICABLE SPECS.	

OPN. NO.	DEFECT CLASS	INSPECTION DESCRIPTION	TOOLS AND GAGES	INSP. STAMP
		b) Monitor oven temp. once every 8 hrs. (continued)		
		48 Hrs. - 32°		
		56 Hrs. - 32°		
		64 Hrs. - 32°		
		72 Hrs. - 31°		
		c) Plates observed and colonies counted after 24 Hrs.,		
		48 Hrs., and 72 Hrs.		
		Count @ 24 Hrs - time & date. 17:00 1-4-67		
		Count " 48 " " " 16:30-1-5-67		
		Count " 72 " " " 16:30-1-6-67		
		d) Log count on data sheets Yes		
5		Log on attached sheets any deviations from procedure.		
		Deviation Log		

REV.					PLANNER	DATE
DATE & INIT.					[signature]	1-3-66

FORM 1557 REV. (4-65)

APPENDIX F

VOYAGER MARS PLANETARY QUARANTINE:

LITERATURE SEARCH - COLD GAS SYSTEMS

BY
J. A. Mason
Propulsion Engineering
Advanced Interplanetary Programs

APPROVED R. P. Wolfson

R. P. Wolfson, Cognizant Engineer
Planetary Quarantine
Voyager Spacecraft System Project

PREPARED FOR

Jet Propulsion Laboratory
California Institute of Technology
4800 Oak Grove Drive
Pasadena, California

Under JPL Contract No. 951112,
Modification No. 9

SUBMITTED BY

Missile and Space Division
Valley Forge Space Technology Center
P. O. Box 8555, Philadelphia 1, Penna.

GENERAL ELECTRIC
MISSILE AND SPACE DIVISION
[illegible]

PROGRAM INFORMATION REQUEST/RELEASE

*CLASS. LTR.	OPERATION	PROGRAM	SEQUENCE NO.	REV. LTR.
PIR NO.	6260	08		

"USE "C" FOR CLASSIFIED AND "U" FOR UNCLASSIFIED

FROM	TO
J. A. Mason Propulsion Engineering/Rm. U3141 X2746	DISTRIBUTION

DATE SENT	DATE INFO. REQUIRED	PROJECT AND REQ. NO.	REFERENCE DIR. NO.
10/19/66		VOYAGER PQ	

SUBJECT

VOYAGER PLANETARY QUARANTINE LITERATURE SEARCH - COLD GAS SYSTEMS

INFORMATION REQUESTED/RELEASED

This literature search was conducted, as part of the Planetary Quarantine Program, to gain data on -- 1) internal contamination levels of cold gas systems, 2) the effectiveness of various techniques used to control biological contamination. The information gained serves as a guideline in determining the effort required to meet quarantine cleanliness requirements and also provides preliminary data to the PQ math model for evaluation.

SYSTEM CONFIGURATION

A typical Attitude Control System (ACS) is composed of propellant tank or tanks, a filter, regulator, solenoid valves and thrusters (for each axis) and associated tubing. The internal area of the Voyager ACS is approximately 26 ft^2. The internal areas are distributed such that 21 ft^2 are in the tanks, approximately 3 ft^2 in the tubing, and the remaining 2 ft^2 for the component internal areas.

EXPECTED CONTAMINATION LEVELS

GE, under NASA contract (Reference 1), has performed an investigation to develop a manufacturing and assembly procedure for planetary spacecraft to be sterilized by heat (GE Report [illegible]). In this investigation, two tests were conducted to determine the biological loading of hardware components after,(1) typical cleaning processes for pneumatic and hydraulic components, (2) cleaning by nitrogen purge. In Test (1), three sets of pressure gages, valves, pressure regulators, and 1/4 in. aluminum tubing 12 inches long were cleaned at the NASA Marshall Space Flight Center in accordance with specific cleanliness specifications.* Another set of like components were evaluated in the "as received" condition. The bioassay results of the test are as follows:

*MSFC - Spec - 164 cleanliness of components for use in oxygen, fuel, and pneumatic systems

*MSFC - Proc - 166 process for cleaning, testing, and handling of space vehicles hydraulic systems components and hydraulic fluids.

*MSFC - 10419906 cleanliness levels, cleaning and inspection procedures for components parts of gas bearing and slosh measuring systems.

DISTRIBUTION:

R. Clapper	F. Mayer
A. Cohen	A. Coats
H. [illegible]	R. Wolfson
H. Leech	

PAGE NO. 1 OF 6

Attach.

Cleaning Method	Surface	Average Bacteria	Microorganisms Recovered/ft^2 Fungi	Spores	Total
None (as rec)	External	4057	8	288	4130
	Internal	2342	164	840	2506
MSFC* Proc-166c	"	288	20	288	308
	"	172	0	96	172
MSFC* Spec-164	"	185	0	120	185
	"	195	0	192	192
MSFC* 10419906	"	132	0	110	132
	"	160	0	160	160

(*See previous page of this PIR)

Over eighty sample points were taken during the test to establish the above results.

Test (2) was performed using two solenoid valves, a pressure regulator, and four pieces of stainless steel tubing, all artificially contaminated with approximately 10^5 spores. The test items were purged with N_2 gas and sterile water and the effluent collected for bioassay. Results are given in the following chart

Contamination After N_2 Purge

Part Name	Purge Volume (ft^3)	Sample** Number	Organisms Recovered
Valve No. 13	10	(1)	1.9×10^3
	10	(2)	7.0×10^3
		(3)	1.2×10^4
Tube 1-1	10	(1)	$> 10^3$
	10	(2)	< 50
		(3)	7.0×10^4
Tube 1-2	10	(1)	10^3
	10	(2)	1.0×10^2
		(3)	$< 10^2$

**(1) Filter assy after first N_2 purge (2) Filter assy after second N_2 purge (3) Sterile H_2O flush effluent assy. The bacteria was flushed after the N_2 purge.

Chart continued ...

Part Name	Purge Volume (ft^3)	Sample** Number	Organisms Recovered
Valve No. 6	100	(1)	1.0×10^3
	100	(2)	1.0×10^3
		(3)	8.0×10^4
Tube 1-3	100	(1)	4.3×10^3
	100	(2)	1.5×10^3
		(3)	1.6×10^4
Tube 1-4	100	(1)	5.0×10^3
	100	(2)	50
		(3)	8.0×10^3
Regulator	100	(1)	*<10
		(2)	*<50

** See previous page (page 2)
* Natural contamination; all other items were artificially contaminated.

In Test(1) swabbing was the primary internal surface sampling method used. In Test(2) it can be seen that the efficiency of a rinse or purge is at least 10%. When correlating these two tests, it can be concluded that the contamination values (organisms/ft^2) given in Test(1) represents, at a minimum, 10% of the microbiological contamination contained in the cleaned hardware samples. Therefore, on an average, the total microbiological loading of the internal surface of a component would be an order of magnitude greater than the recovered values given in Test(1).

For the Voyager Attitude Control System, which has approximately 25 ft^2 of surface area, the microbiological loading would be approximately 52,000 total microorganisms. This approximation is made assuming that tankage contamination levels are approximately that of the components tested. It is expected that tanks, through careful manufacturing procedure, would provide no greater contamination level than the smaller components.

The biological loading contribution by Nitrogen gas used in an ACS is thought to be minimal. JPL, in their Venture Scout Spacecraft, utilized membrane filters in the inlet manifold for biological contamination control. The effectiveness of this filtration has not been reported, however, manufacturers of membrane type filters claim over 99% removal of organisms under carefully controlled evaluation tests.

PERCEPTAMINATION METHODS

Sterilization by Heat. The application of heat is probably the most positive means of securing sterility. Heat sterilization is usually performed in a steam autoclave or by the dry heat method A heat cycle of 135°C for 24 hours would lower the number of viable resistant microbes by a factor of 10^{12} (Reference 2). This would bring the microbiological loading in the ACS (10^4) to a probability of 10^{-8}, if the process could be applied. Preliminary study has indicated that certain components currently used in an attitude control system, particularly those with elastometer parts, O rings, seals, etc., would not be able to withstand the sterilization time temperature cycle and maintain their reliability. However, the biological kill by heat sterilization follows a log rate. Therefore, the kill probability would be reduced by reducing the temperature and/or time of the sterilization cycle. The reduced temperature and/or time cycle would have less adverse effects on component reliability and function Charts "b" and "c" of attachment I (NASA Approved Procedure) show time and temperature for bacterial population decrease of 4 to 10 decades. The Martin Co. is currently under contract to JPL to conduct a design and experimental investigation and a feasibility demonstration of a heat sterilized propulsion system Results of this investigation should provide information on component sensitivity to the sterilization temperature

Decontamination by Gases There are several gases that have been proven to be lethal to viable spores. Of these Ethylene Oxide has been used most extensively, and provides most of the data. Ethylene Oxide (EtO), as with all gases, decontaminates exposed surfaces only. Sealed areas and stagnant areas are not readily reached and consequently, may escape decontamination. On surface areas, where appropriate concentrations of EtO can be applied under control temperature, and humidity conditions, it has been shown to kill 10^6 resistant organisms (References 3). Normal time for EtO exposure is 8 to 18 hours. Longer exposure has not shown to produce further microbiological population decrease. It is anticipated that materials can be selected for ACS components that will withstand EtO environment and maintain performance integrity.

Decontamination by Liquids. As with gas, liquid decontamination only cleans exposed surfaces, but because of the higher viscosity and surface tension of liquids, it will not reach into crevice and crevices as readily as a gas. Liquid decontaminants usually require less contact time than that of gases. Tests to date have shown liquid decontaminants to provide a kill rate equal to that of gas under appropriate control conditions of concentration, quality and temperature (References 4). The formaldehydes (40% aqueous solution-Formalin) and hypochlorites are the more commonly used liquid sterilants.

Gas Decontamination by Filtration. Filtration has been proven to be an effective means of removing viable spores and/or bacteria, from liquid and gaseous fluids, in the pharmaceutical, medical, and hospital fields. Various filters of uniform pore size down to .02 microns are currently available in the industry. Nine one hundred percent removal of bacterial spores are claimed by various manufacturers based on carefully controlled testing Further study is currently being performed by Pall Corp. Gelman Company, under JPL contract, to evaluate "sterilization filters" in a propulsion gas flow system. This study will also provide a more accurate means of evaluating filter efficiency and reliability.

COMPONENT INTERNAL SURFACE DECONTAMINATION

Very little information has been found on the decontamination of component and/or system internal surfaces. The most significant work was that performed by GE in Reference 1. This effort was primarily the bioassay of components after standard methods of cleaning.

Areas concerning component internal surface contamination requiring further investigation are:

1. Effects of system assembly and/or component replacement on bioclealiness.
2. Effectiveness of ETO purge or liquid decontaminant flushes.
3. Efficiency of bioassay techniques.
4. Load reduction by gas purge.

STERILIZATION PROCESS

The major space programs that required quarantine are the Ranger, Mariner Series, and the Surveyor Spacecraft. The quarantine program for the Ranger Series of spacecraft required a heat cycle of 125°C for 24 hours for those parts that could take the sterilization temperature. Subsystems that could not be heated were divided into heatable and non-heatable components. The heatable components were subjected to the temperature cycle, and the heat sensitive components were assembled to the heated portion in a dry box after all systems were subjected to ETO. The main assembly procedure (assembly of subsystems to spacecraft structure) used isopropanol to clean surfaces before mating.

From available information, it appears that the Mariner Series used essentially the same sterilization procedure with exception to the sterilization temperature, which was raised from 125°C to 135°C.

During the Ranger and Mariner program, many waivers were granted to parts that could not withstand the sterilization environment. No bioassay values, actual or calculated, for the Ranger-Mariner decontamination procedure were found during the literature search.

A quarantine program was also required for the Surveyor Spacecraft. No applicable information on this program was found.

CURRENT STERILIZATION ACTIVITIES

The following current activity items were selected from JPL program and technical reports, and will be monitored on the way effect biological decontamination of AMS efforts:

1. Biological stability indicator for dry heat sterilization. JPL - A. Ennis
2. Microbial filters for liquid and gas. JPL - A. Ennis/Millipore Corp.

3. Sterile assembly techniques. JPL/Lockheed

4. Ethylene Oxide sterilization study. JPL - L Reed

5 Pressure transducer sterilization. JPL - C A Crawford

6. Sterilization of polymeric material JPL - H Harvey

DISCUSSION

An attitude control system of the size considered for Voyager, that has been manufactured under current cleanliness standards and assembled in at least a class 100,000 clean room, can be expected to have from 10^4 to 10^5 viable organisms on the interior surfaces. The number of organisms discharged by firing a system has not been determined. Component purging with N_2 gas has been shown to decrease the component microbiological loading by an order of magnitude. A comparison between a purge test and actual flight conditions cannot be made with confidence since vibration and cyclic operation effects upon the quantity ejected are unknown However, as a worst case, it could be considered that the microbes are ejected in proportion to the propellant usage. This value could be used as input to the math model as a worst case consideration. Should this value not exceed planetary quarantine restraints it could be concluded that the currently available cleanliness standards and procedures are adequate. Should this ejecta value exceed the quarantine restraint, then there seems to be three primary alternatives:

1. Evaluate an ACS system to more accurately determine ejecta discharge loading.

2. Decontaminate by EtO, or other non-heat decontamination processes

3. Heat sterilize the system

The evaluation of the ACS ejecta discharging loading would provide more precise data which could be used in place of the worst case consideration The actual ejecta bioloading may be sufficiently low to be within quarantine limits System ejecta potential could also be reduced by placing a bio-filter (membrane) upstream from the regulator. This filter would isolate 99% of the system internal surface area (tank used) and consequently reduce the system ejectable bio-loading by an order of magnitude. System performance would not be effected by use of the filter. Filters with this capability are currently available in the industry. Should the quarantine restriction on ejecta be sufficiently severe that filtration and cleaning would not satisfy the requirement, then EtO or heat sterilization would be required.

REFERENCES

Reference 1 - NASA Contract No. NAS 8-11372

Reference 2 - C. W. Brech, Dry Heat Sterilization of Components for Space Probes. Wilmot Castle Co. Status Report No. 2, NASr-31

Reference 3 - L. D. Jaffe, Sterilization of Unmanned Planetary and Lunar Space Vehicles. JPL Report No. NSS-18430, 1962

Reference 4 - J. B. Opfell, Evaluation of Liquid Sterilants, Dynamic Sciences Corp. Final Report, Contract No. N-2-150747.

As of July 1966, only dry heat has been approved by NASA for the sterilization of planetary landers. Reference 3-2, issued on 21 July 1965, specifies the 12 heat cycles listed in Table a. The D values shown in the table are based on heterogeneous mesophilic bacterial spores in soil (Ref. 3-3). The specified cycles will reduce the bacterial population by 12 decades; i.e., from 10^6 to 10^{-4}.

Decontamination, i.e., a reduction in bacterial population without complete sterilization, may be achieved by any of the heat cycles of Tables b or c depending upon whether or not the hardware item can be satisfactorily bioassayed, (Ref. 4.22-2). The D-values for Table b are based on spores of B. subtilis var. niger in plastics and the stated times achieve a 4-decade reduction. The D-values for Table c are based on spores in soil and the times achieve a 10-decade reduction. Surfaces may be decontaminated by ethylene oxide as specified in Table d.

a

TEMPERATURE (°C)	STERILIZATION TIME (HR)	D VALUE (HR)
163	3	0.33
155	6	0.51
150	6	0.43
145	9	0.79
140	24	1.3
135	32	1.8
130	48	2.6
125	60	4.4
120	61	7.0
115	100	11.0
110	120	17.8
105	200	32.0

APPROVED STERILIZATION CYCLES

b

TEMPERATURE (°C)	D VALUE (HR)	DECONTAMINATION TIME (HR)
163	0.4	0.60
155	0.6	0.96
150	1.5	2.30
145	4	3.0
140	7	5.7
135	10	8.5
130	17	15
125	120	23

APPROVED DRY HEAT DECONTAMINATION CYCLES FOR ASSAYABLE ITEMS

c

TEMPERATURE (°C)	D VALUE (HR)	DECONTAMINATION TIME (HR)
163	2.0	22.0
160	2.5	27.5
155	2.0	22.0
150	1.0	9.0
145	0.6	6.6
140	0.8	8.8
135	0.3	3.3
130	0.2	2.2
125	0.13	0.52
120	0.06	0.60
115	0.04	0.44
110	0.03	0.33

APPROVED DRY HEAT DECONTAMINATION CYCLES FOR NONASSAYABLE ITEMS

* Extracted from

NASA Procedures Manual for Planetary Spacecraft to be Sterilized by Heat (July 5, 1966) (Vol. II) NASA Contract No. NAS 8-11372 to GE, S/D, KSC.

APPENDIX G

VOYAGER MARS PLANETARY QUARANTINE:

COLD GAS ATTITUDE CONTROL SYSTEMS,

EXPERIMENTAL PROGRAM - STATUS

REPORT, JANUARY 1967

PREPARED BY:

J. A. Mason
Propulsion Engineering
Advanced Interplanetary Programs

APPROVED: R.P. Wolfson

R. P. Wolfson
Cognizant Engineer
Planetary Quarantine
Voyager Spacecraft System Project

PREPARED FOR:

Jet Propulsion Laboratory
California Institute of Technology
4800 Oak Grove Drive
Pasadena, California

Under JPL Contract No. 951112

GENERAL ELECTRIC
Missile and Space Division

- STATUS REPORT -

A. SUMMARY

This report presents the work performed to date in the design, fabrication, cleaning and testing of a cold gas (Nitrogen) attitude control system to determine biological content of the discharge gas. The system was designed to be functionally similar, utilize typical ACS hardware, and to have its internal areas proportional to a Voyager-size attitude control system (ACS).

The ACS consist of a propellant tank, fill valve, filter, regulator, pressure transducers, solenoid valve, thruster and associated tubing and fittings. All components and the system were cleaned in conformance with the Nimbus Satellite cleanliness specifications and assembled in a clean room. The system was activated on a duty cycle consisting of short and long pulses and blowdown periods. The discharge gas from each firing was filtered through a 0.45 micron membrane filter. Each filter element was then bioassayed to determine ejecta bioloading. Testing was also conducted to determine the effects of vibration on biodischarge.

B. INTRODUCTION

The study to determine the effects of propulsion systems on the maintenance of Mars planetary quarantine is being conducted as part of the Voyager Task C Planetary Quarantine Study Program. A potential source of contamination is the release of viable organisms from the spacecraft cold gas attitude control system. The cold gas system study is being conducted in two phases, 1) a literature search, to gain data on the effectiveness of various biological control techniques, and the quantity of biological contamination that can be expected to

be ejected from an ACS, and 2) a test program to determine the quantity of organisms discharged from an ACS that has been cleaned under standard or normal procedures. The literature search was completed in October, 1966, and is reported in detail in Appendix F. . Data obtained from the literature search revealed that hardware components, after specified cleaning, can be expected to contain approximately 2000 viable organisms (spores) per square feet of internal surface, and that purging of hardware components with nitrogen gas was relatively ineffective in reducing the internal contamination level. No work was found that evaluated the ejecta of attitude control systems that were cleaned and assembled under "normal" spacecraft pneumatic system procedures. Consequently, the testing conducted in this phase (Phase 2) of the propulsion quarantine task is directed towards the determination of the quantity of viable spores ejected and the rate of such ejection.

C. PROGRAM TEST OBJECTIVES

The ACS ejecta bioassay is being performed to determine:

1. The viable organisms ejected from a typical cold gas ACS that has been cleaned and assembled under "normal" (currently used) procedures.
2. The rate of bio-ejection under pulse and steady state modes of system operation.
3. The quantity of organisms ejected at intervals along the duty cycle.
4. The effect of vibration on organisms ejection rate. The applied vibration would be typical of that expected for Voyager.

D. SYSTEM DESIGN AND OPERATIONAL CONSIDERATIONS

The system was designed to provide ejecta results that could be used to predict, within reasonable accuracy, the quantity of viable ejecta from an actual Voyager ACS, along its flight path. The following design considerations were applied.

1. The ratio of propellant tank internal area to all other component and line internal areas is the same for the test system as for the Voyager ACS proposed by G.E. in the Task B design. This ratio is approximately 20/80 and represents the high and low velocity gas areas.
2. The test system utilizes typical ACS pneumatic components, to give representative internal areas and flow passage configurations.
3. A thruster to provide the same system gas flow rate as any single thruster on the Voyager System.
4. Provide a test cycle containing short and long duration pulses at intervals along the total ACS operational spectrum.
5. Vibration to be applied to the test system at the expected Voyager frequencies and accelerations prior to system operation to determine the effect of vibration on bio-ejection.
6. The system to be pressurized with bio-filtered gas (filtered through .45 micron filters) so that all ejected organisms may be considered to be removed from the internal surfaces of the system.

E. SYSTEM AND HARDWARE DESCRIPTIONS

The system layout is given in Attachment 1, G.E. drawing number SK 56152-697. The system is conventionally designed and comprised of the following components: Propellant Tank, pressure transducers (high and low pressure), regulator, filter solenoid valve, thruster and associated tubes and fittings. The components are rigidly mounted on a 3/4 in. aluminum plate so the system will not be damaged

during vibration testing. An adapter plate is mounted adjacent to the thruster to hold the ejecta filter. "O" ring seals are used on both faces of the adapter to prevent gas leakage during system operation. Hardware used in the system was selected from components used during development of the attitude control pneumatic systems of Nimbus, Advent, and OAO. These components have been subjected to normal handling and storage contamination associated with a development program, with the exception of the propellant tank which is a new item designed for the Gravity Gradient II Satellite. One-quarter-inch-diameter, 0.035 inch wall, 304 stainless steel tubing, in conjunction with MS flared fittings, were used to connect the components into a system.

Figure 1 shows the total system test schematic. Pictures of the system as tested with ejecta filters in place are given in Figures 2a and 2b. Table 1 provides a brief identification of the primary components and Table 2 gives the system primary operating parameters.

Table 1. Component Description and Usage

COMPONENT	PROGRAM	MANUFACTURER	INTERNAL AREA
1. Propellant tank - 145 $in.^3$	GG II	Airite	142 $in.^2$
2. Check valve -	OAO	Circle Seal	2.0
3. Filter - 10 microns	Advent	Airc Porous Media	7.4
4. Transducer (LP) - 30-40 psi	Nimbus	Trans-Sonic	0.5
5. Transducer (HP) - 0-3700	Nimbus	Trans-Sonic	0.5
6. Solenoid valve - 1/4"	Advent	Valcor	3.0
7. Regulator - 30 psi Reg. press	Nimbus	Carlton	11.0
8. Thruster - 0.1# thrust	Nimbus	G.E.	1.5
9. Lines - 1/4 dia. .035 wall			10.6

Table 2. System Operating Parameters

Tank Volume	145 in^3
Tank Pressure	2000 psi
Pounds of gas	0.885 lb
Regulated Pressure	39 psi
Thruster Chamber Pressure	36 psi
Gas Flow Rate	0.0019 lb/sec
System Thrust	0.1 lb
System Total Internal Area	178.0 in^2
Tank to System Area %	79.5%

F. CLEANING AND ASSEMBLY PROCEDURES

The Nimbus cleaning and assembly procedures were chosen as typical. These pneumatic systems specifications are given in Appendix C.

In general, all hardware components are disassembled in the flow region, inspected and cleaned. The cleaning is to G.E. Spec. No. SVS 2631E Para. 3.7 (Appendix C). The degreasing referenced in the cleaning spec consists of ultrasonic cleaning the disassembled components in a freon bath. Each component is purged after reassembly and a particulate count taken of the effluence. Purging is continued until the specified cleanliness level is reached. After cleaning the component is sealed in clean plastic bags. Subsequent functional testing of the components are performed with test fluid that has been filtered to a .45 micron level. System cleaning and assembly are performed in accordance with Spec. SVS 2631E and G.E. Quality Control & Test Standing Instruc. No. S1-236659 Appendix C. The assembly work was performed in a newly constructed clean room that was designed to meet Class 100,000 cleanliness level. The actual cleanliness level has not been established through measurement; however, cleanroom maintenance and practices are being observed. Final system acceptance is based upon meeting particle count as given in SVS 2631E Para. 3:7.4.

G. EJECTA FILTRATION

The discharge gas from the ACS is fired into a Gelman Instrument Co. filter holder (PN 11101) containing a .45 micron membrane filter. Evaluation testing of the Gelman filter holder and the method of attachment to the thruster was made on an auxiliary ACS. The auxiliary system utilized a thruster and solenoid valve of the same configuration as that used in the test ACS. The auxiliary system was pressurized from precharged gas cylinders and regulated by an oxygen welding regulator. The design of the filter holder to thruster adapter method, techniques to prevent contamination of the sterile filter holders during testing and procedures for removing the membrane filters from the filter holders for bioassay were developed during testing of the auxiliary system. Filter holder $\triangle P$ was also determined to assure that supersonic flow would be maintained in the thrust of the thruster during testing.

H. TESTING AND BIOASSAY PROCEDURE

The testing and bioassay procedures are as given in Appendixes A and D, respectively. Quality control provided data and procedural checks for monitoring and recording of each step of the testing, cleaning and bioassay.

I. TEST PLANS

The ACS test plan is as follows; variations to the test plans may be prudent as the testing progresses.

TEST X

Step 1. Three pulses of 5 second duration each.

" 2. Three pulses of 30 ms duration each.

" 3. System blowdown to 50% pressure.

" 4. Repeat step 1.

TEST I (Cont.)

Step 5. Repeat Step 2.

" 6. System blowdown to 25% pressure.

" 7. Repeat step 1.

" 8. Repeat step 2.

TEST II

Step 1. System vibrated to simulate boost phase.

" 2. Three pulses of 5 seconds duration each.

" 3. Three pulses of 30 ms duration each.

" 4. System blowdown to 50% pressure.

" 5. Repeat steps 2 and 3.

" 6. System blowdown to 25% pressure.

" 7. Vibrate system to simulate orbit insertion firing.

" 8. Repeat steps 2 and 3.

TEST III

Step 1. Repeat Test I.

TEST IV

Step 1. Repeat Test II.

A new ejecta filter is used after each system pulse. In addition to the above testing, a bioassay of system components is to be performed after all system testing is completed.

J. TEST RESULTS

Test I and II have been completed to date. Data from Test (I) is given in Table 3.

TABLE 3

STEPS	FLOW TIME sec.	GAS VOL. $(ft)^3 \times 10^{-2}$	FILTER NUMBER	AVERAGE NUMBER OF VIABLE ORGANISMS/MEMBRANE FILTER NON-HEAT SHOCKED	HEAT SHOCKED
1	5.7	1.04	1	0	< 5
	4.7	.89	2	< 10	0
	5.8	1.05	3	< 5	0
2	.28	.053	4	0	0
	.3	.057	5	0	< 5
	.39	.074	6	0	< 5
3	180.00	34.2	7	< 10	0
4	5.8	1.05	8	< 30	0
	5.6	1.03	9	< 5	< 5
	5.9	1.06	10	< 10	0
5	.42	.08	11	0	0
	.32	.06	12	< 10	0
	.42	.08	13	< 5	0
6	124.00	23.5	14	0	0
7	5.2	1.0	1	< 20	< 30, > 20
	5.25	1.0	2	0	0
	5.1	1.0	3	< 10	0
8	.35	.065	4	< 30	0
	.26	.05	5	< 10	*
	.55	.01	6	0	*
9	80.00	15.1	Blank	< 5	*

Fallout 0.367 organisms/petri dish-min. or
0.00467 organisms/cm^2-min.

*discarded because contaminated during one of the procedures.

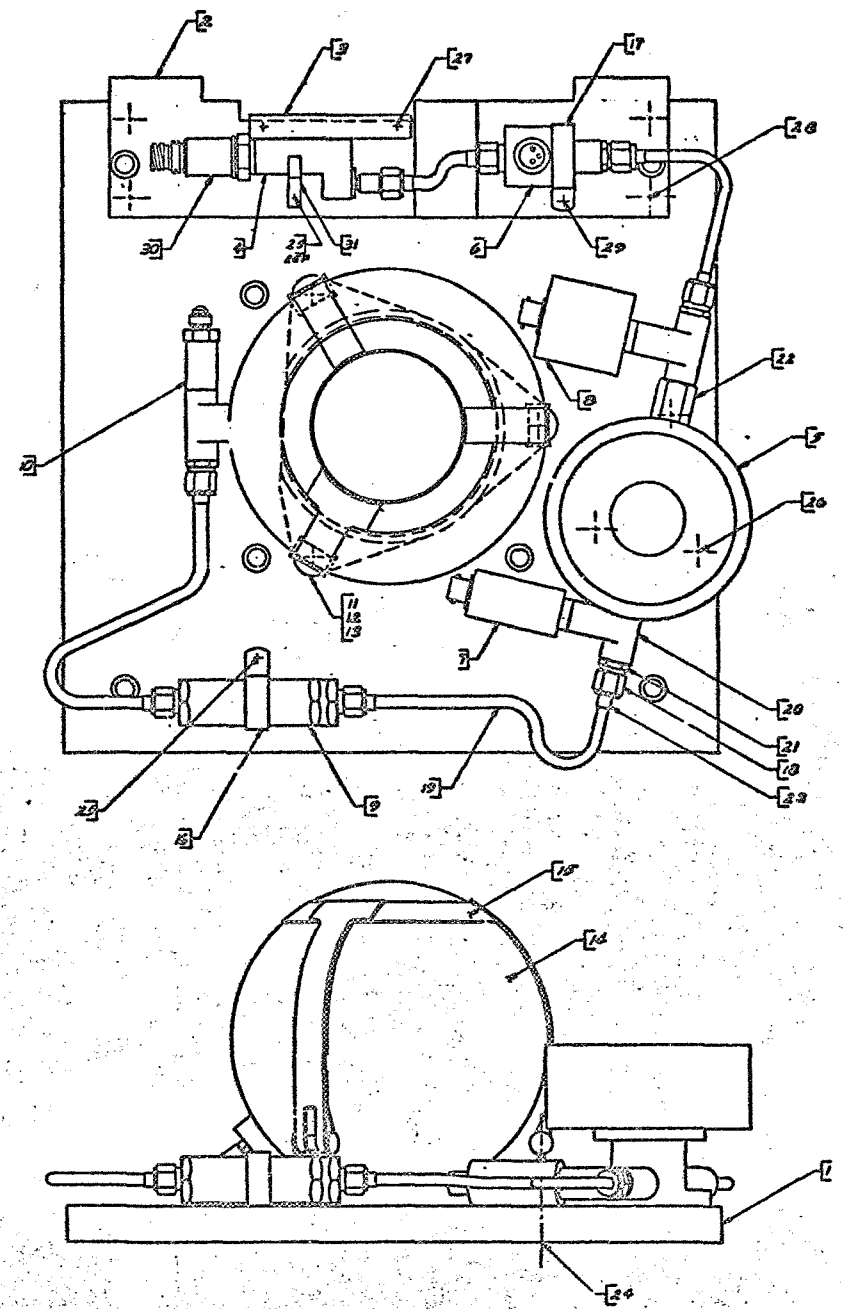
The data shows that 13 of the 18 valid bioassays (three were inadvertently contaminated) did not contain any viable spores. Of the remaining filter elements, only one (filter #9) had evidence of being contaminated by the ACS system. Filter elements 1, 5, 6 and 1(Step 7) provided inconsistent results, i.e., greater contamination observed after heat shock, which indicate contamination during the bioassay procedure. In general, the number of organisms recovered are of such a low value that statistical method normally applied to determine the ejected quantity are not valid. Biological laboratory background contamination obscure such low bioassay results. In consideration of the low contamination level obtained in Test I, the ejecta filtration procedure was revised for Test II to allow more gas to flow through a filter for a particular system test. The revised procedure allows two pulses to be passed through a filter before the filter is changed, and also revises the number of pulses in a test. The revised test procedure is given in Appendix A. During the initial vibration portion of Test II a leak was developed which bled the system down to 1300 psi before the system was activated for Test II. Only Steps 1 through 5 of Test II were completed before the tank pressure decreased below the value required for a second sequence. The remaining gas was blown through a single filter.

Preliminary indications (72 hour count) reveals very low spore count, i.e., less than 10 spores for the total test. Actual gas quantity and system "valve on time" have not been reduced from recording traces at the time of this report.

As a check on system internal contamination level the in-line 10 micron filter unit was removed from the system after Test I and a bioassay of the filter internal surfaces was performed. The test results after 72 hours incubation show no viable organisms in the culture.

K. PLANNED ACTIVITY

1. As a result of the very low number of viable organisms being ejected in the two tests that have been performed, it is now planned only to run one additional vibration test series to verify previous results. The bioassay will be altered to that given in Appendix D. This appendix, in turn, provides a bioassay procedure for use when it has been previously established that less than 30 organisms are being deposited per filter element. This procedure minimizes biological laboratory contamination.
2. Test the ejecta of a flight or engineering model ACS that has been manufactured in house (G.E.) under cleaning processes similar to that of the Nimbus. This test would be desirable to verify results of the scaled ACS test.
3. Perform bioassay on all components of the test ACS system to determine interior surface contamination.
4. Determine propellant gas microbiological loading.



ATTITUDE CONTROL,
PNEUMATIC SYSTEM

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